The conversion of circular genomes to linear chromosomes during molecular evolution required the invention of telomeres. This entailed the acquisition of factors necessary to fulfill two new requirements: the need to fully replicate terminal DNA sequences and the ability to distinguish chromosome ends from damaged DNA. Here we consider the multifaceted functions of factors recruited to perpetuate and stabilize telomeres. We discuss recent theories for how telomere factors evolved from existing cellular machineries and examine their engagement in non-telomeric functions such as DNA repair, replication, and transcriptional regulation. We highlight the remarkable versatility of protection of telomeres 1 (POT1) proteins that was fueled by gene duplication and divergence events that occurred independently across several eukaryotic lineages. Finally, we consider the relationship between oxidative stress and telomeres and the enigmatic role of telomere-associated proteins in mitochondria. These findings point to an evolving and intimate connection between telomeres and cellular physiology and the strong drive to maintain chromosome integrity.

Molecular evolution is opportunistic, enabling novel cellular mechanisms to arise in response to biological challenges. One such challenge was conversion of the circular prokaryotic genome into the multiple linear DNA forms that comprise the eukaryotic genome (1). This challenge necessitated the invention of telomeres. Here we discuss the origin and evolution of telomere-related functions. Although the factors associated with chromosome ends were initially thought to be specific for this locale, in-depth analysis has revealed many such factors having noncanonical, so-called “moonlighting” roles in other transactions within the nucleus and the cytoplasm. We now appreciate that some of the moonlighting contributions may reflect ancestral functions preserved from the dawn of genome linearization, whereas others may be newly emergent.

There are several theories for how linear chromosomes evolved from their circular progenitors (2, 3), but one of the more intriguing proposals is that invasion of circular genomes by group II introns (1), via reverse splicing and reverse transcription, led to DNA linearization (4, 5) (Fig. 1). Specifically, it is posited that non-LTR2 retrotransposons targeted to double-strand breaks (DSBs) served as “proto-telomeres” (6). The nascent chromosome ends presented two immediate challenges: the “end replication” problem and need for “end protection” (7, 8). The end replication problem occurs because the DNA replication machinery cannot fully replicate the extreme terminus of the lagging strand, which would lead to the gradual depletion of terminal DNA sequences when the genome is duplicated (9, 10). The chromosome ends may also be perceived as a DSB and must therefore be sequestered to prevent activation of the DNA damage response. Such end protection is also crucial for the avoidance of end-to-end fusions of chromosomes, which would cause improper chromosome segregation during mitosis, cell cycle arrest, genome instability, senescence, and cell death (8, 11). Most eukaryotes cope with these problems by 1) adding long arrays of noncoding DNA repeats to serve as a physical buffer to protect coding regions from attrition and 2) formation of higher-order DNA architecture that helps distinguish chromosome ends from a DSB (i.e. fold-back structures in yeast (12) and t-loops in other species (13, 14)).

Emergence of telomerase

To help overcome the telomere end replication problem, a group II intron likely gained the ability to use the 3’ end of linearized chromosomes as a template for reverse transcription (5). There is strong evidence that the telomerase catalytic subunit TERT evolved from a non-LTR class 2 retrotransposon (15–17) (Fig. 1). Fruit flies and silkworms maintain their chromosome ends through a telomerase-independent mechanism that employs a different class of retrotransposons (18, 19), supporting the idea that retrotransposons played an early and critical role in establishing and maintaining telomere architecture (20, 21).

The modern-day enzyme that helps solve the end replication problem is telomerase, a reverse transcriptase that compensates for incomplete replication by continually replenishing terminal DNA using a long noncoding RNA, TER, as template (22). It is possible that TER arose from a transcript derived from the progenitor group II intron (5) (Fig. 1), but TER and TERT

2The abbreviations used are: LTR, long terminal repeat; DSB, double-strand break; NHEJ, nonhomologous DNA end joining; OB-fold, oligonucleotide/oligosaccharide-binding fold; ROS, reactive oxygen species; 8-oxoG, 8-oxo-guanine; Tg, thymine glycol; BER, base excision repair; MTS, mitochondrial targeting sequence.
Telomere-associated proteins: Origins and their role in telomere end protection

In vertebrates and fission yeast, telomere end protection is mediated by shelterin (29, 30) (Fig. 2 and Table 1). Shelterin physically caps the telomere ends, preventing the termini from being recognized as DNA damage and suffering DNA attrition via nucleolytic processing and DNA damage checkpoint activation. Shelterin is composed of TRF1/TRF2 (SpTAZ1), which binds the duplex DNA, and POT1-TPP1/SpPot1-SpTpz1, which binds the 3′-single-strand extension on the extreme terminus (termed the G-overhang). Additional proteins bridge the two DNA-binding complexes (TIN2/SpPOZ1) and RAP1. In addition to end protection, shelterin controls telomerase access and therefore contributes to telomere length regulation (29).

In budding yeast, instead of chromosome end protection by shelterin, the G-overhang is stably bound by the CST complex, comprised of Cdc13(CTC1), STN1, and TEN1 proteins (31) (Fig. 2 and Table 1). Notably, vertebrates also possess CST, but this complex only transiently associates with telomeres during S phase to promote telomeric DNA replication. CST is structurally related to the single-strand DNA-binding complex RPA and had likely evolved from the latter (32, 33). Interestingly, Drosophila lacks canonical telomere repeat arrays at its chromosome termini and yet encodes one or more proteins related to CST subunits (34, 35). Flowering plants, including Arabidopsis, present yet another twist on the telomere protection apparatus wherein one half of the chromosome ends harbor a G-overhang bound by CST, whereas the other half are blunt-ended and bound by Ku, which functions in the nonhomologous DNA end joining (NHEJ) pathway of DSB repair and has high affinity for DNA ends (36) (Fig. 2). The asymmetry of plant telomeres may reflect the absence of a 5′ exonuclease (e.g., Apollo) (37) that normally converts blunt-end telomeres created from leading-strand synthesis into termini with the typical 3′ G-overhang (38).

The shelterin and CST proteins employ one of two DNA binding motifs: the MYB domain for duplex DNA binding and the oligonucleotide/oligosaccharide-binding fold (OB-fold) for interaction with single-strand DNA. The MYB motif is common in transcription factors and may have been predisposed to function at telomeres as it is capable of binding tandemly repeated sequences (39, 40). OB-folds, on the other hand, function in a vast array of nucleic acid transactions and are found in proteins ranging from t-RNA synthetases to nucleases and RPA (41). Thus, the single-strand telomere-binding proteins have likely diverged from a common OB-fold ancestor with a role in DNA repair and/or replication (42).

Telomere protection and DNA repair

DNA damage repair pathways and telomere-associated proteins act collaboratively to promote genome integrity. Both TERT and TER have been linked to the DNA damage response (Table 1). In the presence of a DSB, human TERT relocalizes to the nucleolus (43), an outcome that would decrease the proba-
ability of de novo telomere formation at sites of DNA damage. In addition, human cells lacking TERT fail to mount an effective DNA damage response to ionizing radiation (44). Intriguingly, these cells also display altered chromatin structure and fragmented chromosomes, suggesting that TERT plays a role in chromatin reorganization (45). Human TER (hTR) has been proposed to play a TERT-independent role in the response to DNA damage. Inhibition of hTR causes rapid arrest of cell growth, whereas increased hTR, which occurs in response to DNA damage induced by UV light, inhibits the DNA damage

Table 1
Localization and functions of telomere-related components
Tabulated is a summary of published experimental data and in silico predictions for core constituents of the telomerase RNP, the CST complex, and the Shelterin complex. Different functions ascribed to POT1 paralogs from vertebrates, worms, plants, and ciliates are highlighted.

| Component | Localization | Ref. | Role at telomeres | Ref. | Proposed role outside telomeres | Ref. |
|-----------|--------------|------|------------------|------|--------------------------------|------|
| TERT      | Nucleus/nucleoplasm/nucleolar/PLA, bodies/intermediate cytoskeleton/mitochondrial membrane | 158, 159 | Telomere length regulation | 96, 99 | Cell division/base excision repair | 133, 171, 172 |
| TRF1      | Nucleus/nucleoplasm/PM, bodies/intermediate cytoskeleton/mitochondrial membrane | 55, 160 | T-loop formation/telomere bending/represses ATM-mediated DNA damage response | 96 | Regulation of transcription by RNA polymerase II/cellular senescence/apoptosis/cell cycle/DNA damage/base excision repair/replication | 55, 73, 74, 93, 133 |
| TIN2      | Nucleus/mitochondria/perinuclear chromocenter | 57, 138 | DNA damage/mitochondrial oxidative phosphorylation | 167 | DNA damage/mitochondrial oxidative phosphorylation | 138 |
| RAP1      | Nucleus/cytoplasm | 161, 162 | Yeast: binds ds telomeric DNA/negatively regulates telomere length | 90, 173 | Gene silencing/transcriptional regulation/oxidative stress response | 60, 90, 135, 136 |
| TPP1      | Nucleus/cytoplasm | 137 | Connects POT1 to TIN2 recruits telomerase stimulates telomerase processivity | 86, 168 | Coats POT1 localization to nucleus | 137 |
| Vertebrates | Nucleus/cytoplasm | 133 | nPOT1: binds ss telomeric DNA/represses ATM-mediated DNA damage response/telomere length regulation/telomerase recruitment/telomere capping | 163 | DNA duplex unwinding/miotic synopsis/base excision repair/NHEJ/replication | 62, 63, 133 |
| POT1      | Nucleus | 163 | nPOT1a: binds ss telomeric DNA/represses ATM-mediated DNA damage response | 102, 103 | Regulation of GTPase activity/recombination phosphorylation/innate immune response | 104 |
| Worms     | Nucleus | 104 | nPOT1b: binds ss telomeric DNA/represses ATM-mediated DNA damage response | 105 | None | |
| Plants    | Nucleus | 105 | CeOB1: G strand binding/telomere length regulation/telomere capping | 105 | None | |
| Ciliates  | Nucleus | 106 | (MRT1): Telomere replication/necessary for telomerase-mediated telomere repeat addition | 106 | DNA crosslink repair/nucleotide excision repair | 108 |
| Nucleus   | 113 | nPOT1a: binds ss telomeric DNA/represses ATM-mediated DNA damage response | 113 | None | |
| Nucleus   | 117 | nPOT1b: binds ss telomeric DNA/represses ATM-mediated DNA damage response | 117 | Telomerase capping/telomere elongation | 85, 117, 120, 121 |
| Cytoplasm (Predicted) | 110 | TPOT1A: promotes genome stability | 110 | Prevents activation of cell cycle checkpoint | 110 |
| Cytoplasm (Predicted) | 109 | TPOT1B: facilitates de novo telomere formation | 109 | Localizes to sites of chromosome breakage but not to telomeres | 111 |
| Nucleus   | 112 | RTP: telomere replication | 112 | None | |
checkpoint kinase ATR (46). In contrast, loss of TR in mice does not trigger phenotypes distinct from those of mTERT mutants, suggesting that the core RNA and protein components of telomerase act in the same pathways (47, 48).

Shelterin proteins also modulate the DNA damage response (Table 1). TRF2, for instance, prevents ATM-mediated DNA damage signaling at telomeres (49) and also helps recruit various DNA damage response and repair factors, such as ERCC1, Apollo, the MRE11-RAD50-NBS1 complex, helicases BLM and WRN, Ku, and PARP1/2 (50) (Table 1). The recruitment of these factors facilitates telomeric DNA replication, promotes the formation of a single-strand overhang on the chromosome terminus, and ensures that telomeres are properly sequenced to prevent inappropriate recombination or activation of a DNA damage response (51, 52). TRF2 can also associate with DSBs within the body of the chromosome as part of the early response to DNA damage (53, 54). As such, the ability of TRF2 to engage the machineries concerned with the DNA damage response and DNA repair likely promotes genome stability on a global scale. Interestingly, both TRF1 and TRF2 are modified by MMS21, a SUMO ligase likely promotes genome stability on a global scale. Interestingly, both TRF1 and TRF2 are modified by MMS21, a SUMO ligase that has been implicated in the DNA damage response (55), a mechanism germane for telomere maintenance in cancer cells that lack telomerase (56).

Like TRF2, TIN2 and RAP1 associate with chromosome locales other than the telomeres. TIN2 accumulates at nontelomeric regions (57) associated with HP1 (58), a heterochromatin mark that has been implicated in the DNA damage response (59). Moreover, in human cells, RAP1 interacts with noncoding interstitial TTTAGGG repeats present on some chromosomes, raising the possibility that RAP1 helps prevent fragility and recombination at these sites (60).

POT1 has also been implicated in the DNA damage response (Table 1) (Fig. 3). The association of POT1 with the telomeric G-overhang prevents activation of an ATR-mediated DNA damage response (61), and recent studies indicate that human POT1 increases the fidelity of NHEJ at nontelomeric sites (62). Intriguingly, the C terminus of hPOT1 bears structural similarity to a Holliday junction resolvase domain (63), supporting the notion that POT1 affects other facets of DNA metabolism beyond telomere biology.

Ku harbors two subunits (Ku70 and Ku80) and is a core component of the NHEJ pathway (64). Within the context of telomere biology, Ku facilitates telomere protection and telomeric DNA replication (36, 65, 66). Recent studies in budding yeast provide clues for how the DNA repair and telomere protection functions of Ku might be parsed at chromosome termini. Ku harbors two solvent-exposed α-helices on opposite sides of the heterodimer. The surface facing the telomere end is necessary for NHEJ, whereas the inward facing helix is required for telomeric heterochromatin formation (67). In addition to discrete structural boundaries, separation of function can be influenced by cell cycle regulation. For example, the cell cycle regulator CYREN was recently shown to interact with Ku and block NHEJ at telomeres during the S and G2 cell cycle phases (68). Ebrahimi and Cooper (69) have postulated that localization of telomeres within different regions of the nucleus influences a broad range of cellular processes, including meiotic recombina-

tion, chromosome segregation, and gene expression. Hence, in a broader sense, both temporal and spatial regulation of telomeres impact cellular physiology.

A role for telomere-associated proteins in DNA replication and transcription

Given that telomere accessory factors have likely evolved from factors that function in DNA repair, DNA replication, and transcription, it is not surprising that some of the telomere-associated factors also function in the aforementioned processes. Because of the highly repetitive nature of G-rich telomeric DNA and its propensity to form higher-order structures, such as the G-quartet, auxiliary factors are needed to ensure timely and proper replication through telomeric tracts. Notably, both POT1 and TRF2 stimulate the helicase activity of WRN (70, 71), and POT1 has been found to promote G-quartet unwinding by the WRN and BLM helicases (72) (Table 1). TRF2 has been proposed to assist in telomeric replication, and it does so by inducing positive supercoiling in DNA that favors enhanced access by DNA topoisomerases and the Apollo nuclease, enzymes critical for replication (73, 74). Furthermore, TRF2 is also hypothesized to assist in the assembly of the pre-replication complex during telomere replication (75, 76).

The primary function of the CST heterotrimer appears to be in telomere replication (Table 1). Originally identified as a DNA Pol α accessory factor (77), the vertebrate CST complex was subsequently shown to stimulate synthesis of the telomeric C-strand after telomerase extends the G-strand (78–80). CST plays a crucial role in the restart of stalled replication forks at nontelomeric sites (81), and CST mutations lead to genome-wide instability (82, 83). Vertebrate CST only transiently engages telomeres (84), but in budding yeast and in Arabidopsis
Table 1

Table 1

95 to the OB-folds of mammalian POT1 (82, 84, 85). Hence, some of the POT1-TPP1 functions within the context of shelterin (86) may be fulfilled by CST. Indeed, Lue (87) has provided a compelling argument that POT1-TPP1 evolved from CST. The multifunctional nature of CST is further evidenced by the involvement of components of the yeast complex in transcriptional regulation through interactions with RNA polymerase II and the elongation factor Spt5. The interactions of CST with the transcription machinery are thought to help mitigate the consequences of RNA polymerase II collision with replication forks (88). In addition, studies in Arabidopsis have revealed that the CST component TEN1 possesses protein chaperone activity that is activated in response to heat stress (89) (Table 1).

Besides CST, other telomere-associated proteins also influence transcriptional regulation (Table 1). Yeast RAP1 was originally described as a transcriptional regulator at many promoters (90, 91). Human RAP1 modulates NF-κB expression (92), whereas interaction of TRF2 with the promoter of the cyclin-dependent kinase CDKN1a affects its expression (93). TERT has also been reported to enhance the expression of genes such as cyclin D1 (94) and NF-κB (95).

Gene duplication: Refining the landscape of telomere protein function

Gene duplication has fueled protein evolution, including telomere proteins. The duplication event giving rise to vertebrate TRF1 and TRF2 dates back 540 million years ago (96), at the beginning of the chordate lineage (97). The conserved C-terminal MYB domain of TRF1/2 facilitates telomeric DNA engagement, whereas divergent N-terminal domains (98) are important for telomeric DNA length regulation (primarily accomplished by TRF1) (99) or chromosome end protection (TRF2) (100) (Table 1). The Candida clade possesses two copies of the gene that encodes the Cdc13 component of CST (101). The two paralogous proteins, Cdc13A and Cdc13B, are significantly smaller than their counterparts in budding yeast and have overlapping but nonredundant functions in telomere length regulation.

One of the most fascinating outcomes of gene duplication is seen with POT1 (Fig. 3) (Table 1). Here, independent gene duplication events occurred repeatedly throughout evolution. Although humans have a single POT1 protein, mice possess two POT1 paralogs, mPOT1a and mPOT1b, that share 72% sequence similarity (102). Recent studies suggest that both mPOT1a and mPOT1b attenuate ATR signaling at chromosome ends (103). However, mPOT1b uniquely contributes to the regulation of 5’ end resection to form the 3’ G-overhang (103) and may also play a cytosolic role in the innate immunity response (104).

The POT1 isoforms in worms and ciliated protozoa exhibit more profound functional divergence. Caenorhabditis elegans encodes four single OB-fold proteins with structural similarity to the OB-folds of mammalian POT1 (105). CeOB1 binds the telomeric G-rich strand, whereas CeOB2 engages the complementary C-rich strand. Mutation in either of these CeOB genes leads to telomere elongation, providing evidence that their encoded proteins serve as a negative regulator of telomerase (106, 107). The function for CeOB3 is unknown; however, CeOB4 (MRT1) was originally identified in a screen for genes required for germ line mortality as a result of telomere shortening (108). CeOB4 is required for telomerase activity in vivo. Intriguingly, CeOB4 also bears a SNM1 family nuclease domain and has been implicated in both DNA cross-link and nucleotide excision repair (108).

In the ciliates Euplotes crassus and Tetrahymena thermophila, there are two POT1 paralogs (109, 110). The Tetrahymena TtPOT1-encoded protein is essential for telomere length maintenance and prevents checkpoint activation much like the vertebrate POT1 proteins (110). However, TtPOT2 protein does not associate with chromosome ends, but instead localizes to internal sites in macronuclear chromosomes that are destined for developmentally programmed cleavage and de novo telomere formation (111). In E. crassus, the telomere end-binding protein caps chromosome ends (109). Replication telomere protein, the other POT1-like protein, is not associated with telomeres, but rather co-localizes with the replication apparatus as it moves through the macronuclear genome (112). This remarkable observation underscores the strong connection between telomere proteins and the DNA replication machinery.

The plant kingdom is replete with large gene families, arising from both localized gene duplication and whole-genome duplication. It is therefore noteworthy that most POT1 genes in plants are not duplicated. The POT1 gene in the early diverging land plant Physcomitrella patens retains the ancestral functions of binding single-stranded G-rich telomeric DNA and protecting chromosome ends from fusion (113). However, at least two independent POT1 duplications occurred in higher plants, one in the grasses and the other in the Brassicaceae family to which Arabidopsis thaliana belongs (114). There are three POT1 paralogs in A. thaliana, AtPOT1a, AtPOT1b, and AtPOT1c (114, 115). AtPOT1a and AtPOT1b exhibit only 52% sequence similarity. AtPOT1a resembles the mammalian shelterin component TPP1 (86, 116) in that it physically associates with the telomerase RNP and stimulates its repeat addition processivity (84, 117). However, unlike TPP1 (118), AtPOT1a accumulates at telomeres only in S phase (117), indicating that it is not a stable component of the end protection complex. Initially, AtPOT1a was not thought to bind telomeric DNA (119), but a recent study showed that the first OB-fold of AtPOT1a has single-strand telomeric DNA-binding activity (120). Strikingly, the AtPOT1a lineage, but not AtPOT1b, has been subjected to positive selection from an ancestral POT1 protein, leading to enhanced interaction with CST (114). Hence, AtPOT1a appears to have been evolved to be specialized for telomere maintenance through CST interaction. A role for AtPOT1b in telomere biology is not clear. It cannot complement the pot1a mutant (114) and cannot bind telomeric DNA in vitro (120). However, overexpression of the AtPOT1b C-terminal domain leads to massive chromosome fusion (121). Whereas this finding implicates AtPOT1b in chromosome end protection, AtPOT1b probably does so in a manner distinct from the single-copy POT1 proteins from vertebrates and fission yeast. The
third POT1 gene in A. thaliana, AtPOT1c, arose only 5 million years ago as a partial duplication of the AtPOT1a locus. The insertion of a transposon into the promoter of AtPOT1c rendered this gene silent almost immediately after its genesis (122). This finding, coupled with the remarkable functional divergence associated with POT1 paralogs across eukarya, argues that POT1 dosage affects the fitness of organisms, and one or more of the duplicated copies must diverge quickly or be silenced.

**Telomere proteins and their role in the genome-wide response to oxidative stress**

The majority of DNA lesions in mammalian and plant cells can be attributed to oxidative damage (123, 124), and recent data indicate that several shelterin components safeguard the genome against this assault (Fig. 4). Reactive oxygen species (ROS) modifies DNA bases, most commonly resulting in 8-oxoGuanine (8-oxoG) and thymine glycol (Tg) (125). If not repaired, 8-oxoG induces GC-TA transversion mutations as well as single-strand or double-strand breaks, leading to genomic instability (126). Tg is the most prevalent oxidative product of thymine, responsible for 10–20% of ionizing radiation-induced genomic damage (127). Due to their high G-T content, telomeres are a hot spot for oxidative damage (128–130).

Base excision repair (BER) is the most important pathway for removing 8-oxoG and Tg lesions (131). Mice lacking the glycosylase NTH1, which removes Tg via BER, exhibit increased telomere fragility (132). Intriguingly, TRF1, TRF2, and POT1 stimulate BER after oxidative damage (133). Interestingly, 8-OxoG and Tg modifications inhibit telomeric DNA binding by TRF1, TRF2, and POT1 in vitro (134). These observations suggest a feedback loop wherein oxidative damage at telomeres leads to the expulsion of the aforementioned telomere proteins, which then become available to assist in the BER-mediated repair of damaged telomeric bases, so as to enable the re-engagement of shelterin at the chromosome terminus (133) (Fig. 4). Because TRF1 and TRF2 can associate with other genomic locales, they may exert a broader impact in the response to oxidative stress.

Recent data reveal an intriguing response of RAP1 to oxidative stress and other types of DNA damage (135). RAP1 levels decrease in the nucleus and the cytoplasm in response to ROS. Diminished levels of cytoplasmic RAP1 appear to promote apoptosis in aging cells (92) (Fig. 4). Notably, in yeast, shortening of telomeres due to senescence releases RAP1, which then becomes associated with extratelomeric sites. Release of RAP1 from telomeres correlates with the down-regulation of genes encoding core histones and the translational apparatus and up-regulation of genes responsive to senescence (136) (Table 1).

**Genome protection from a distance: The role of telomere proteins outside the nucleus**

The role of telomere proteins in the response to oxidative stress correlates with cytoplasmic activities, but the molecular mechanisms that govern telomere protein function outside the nucleus are largely unexplored. In addition to RAP1, several other telomere-related proteins accumulate in the cytoplasm (Table 1). POT1, TPP1, and trace amounts of TIN2, which promote mitochondrial functions that protect against apoptosis.

Function of telomere-related factors

Figure 4. Impact of oxidative stress on telomeres and telomere-associated proteins in mammals. Telomeres are a hot spot for oxidative damage causing base modifications including thymine to thymine glycol and guanine to 8-oxoG. These lesions interfere with DNA binding by TRF1, TRF2, and POT1. These same proteins stimulate BER at telomeres and perhaps elsewhere in the genome, enabling the removal of damaged bases from the DNA. Oxidative DNA damage decreases the abundance of both cytoplasmic and nuclear RAP1, which in turn triggers apoptosis. Conversely, oxidative stress leads to the accumulation in mitochondria of TERT and TIN2, which promote mitochondrial functions that protect against apoptosis.
TERT in the mitochondria has been proposed to stimulate mitochondrial DNA replication and repair (142). Compared with WT mice, the RNA expression profiles of tert mutants monitored for four consecutive generations (G1–G4) reveal statistically significant changes in the expression of both mitochondrial and nuclear encoded genes required for oxidative phosphorylation, mitochondrial function, and antioxidant defense (146). Similar results were obtained A. thaliana tert mutants of generations G2 and G7 (147). Interestingly, yeast and ciliate TERT proteins lack an MTS (139), raising the possibility that the mitochondrial function of TERT is not conserved in these species or that these TERT proteins are transported into mitochondria via a different mechanism.

Conclusions and future directions

With the advent of linear chromosomes, factors involved in different facets of DNA metabolism were coopted to solve the telomere end protection and end replication problems. Some of these factors retain their functions in DNA replication, DNA repair, and transcriptional regulation. Telomeric DNA is a magnet for oxidative damage, and hence in the drive to maintain genome integrity, telomere proteins may have gained the capacity to protect chromosome ends from this assault by promoting BER proximally, or at a distance by affecting mitochondrial function. Alternatively, some noncanonical functions of telomere proteins may have an older origin. Mitochondria, which possess group II introns (148) and proteins structurally similar to the ancestral OB-folds of RPA (149), emerged 1.45 billion years ago (150). Thus, the building blocks for some of the modern-day telomere proteins and their functions in the oxidative stress response may reflect a mitochondrial ancestry. Finally, the ancient and emerging functions of telomere proteins may have an older origin. Mitochondria, mitochondrial DNA replication and repair (151, 152), and thus in the drive to main-

Acknowledgments—We apologize to colleagues whose work we were unable to cite due to space limitations. We thank members of the Shippen laboratory for insightful comments on the manuscript and the American Society for Biochemistry and Molecular Biology for highlighting our research. Components of all of the figures were created using BioRender software.

References

1. Cavalier-Smith, T. (2010) Origin of the cell nucleus, mitosis and sex: roles of intracellular coevolution. Biol. Direct. 5, 7 CrossRef Medline
2. Ishikawa, F., and Naito, T. (1999) Why do we have linear chromosomes? A matter of Adam and Eve. Mutat. Res. 434, 99 – 107 CrossRef Medline
3. Volff, J.-N., and Altenbuchner, J. (2000) A new beginning with new ends: linearisation of circular chromosomes during bacterial evolution. FEMS Microbiol. Lett. 186, 143 – 150 CrossRef Medline
4. Eickbush, T. H. (1997) Molecular biology: telomerase and retrotransposons: which came first? Science 277, 911 – 912 CrossRef Medline
5. de Lange, T. (2015) A loopy view of telomere evolution. Front. Genet. 6, 321 CrossRef Medline
6. Garavis, M., Gonzáles, C., and Villasante, A. (2013) On the origin of the eukaryotic chromosome: The role of noncanonical DNA structures in telomere evolution. Genome Biol. Evol. 5, 1142 – 1150 CrossRef Medline
7. Lingner, J., Cooper, J. P., and Cech, T. R. (1995) Telomerase and DNA end replication: no longer a lagging strand problem? Science 269, 1533 – 1534 CrossRef Medline
8. de Lange, T. (2009) How telomeres solve the end-protection problem. Science 326, 948 – 952 CrossRef Medline
9. Watson, J. D. (1972) Origin of concatemeric T7 DNA. Nat. New Biol. 239, 197 – 201 CrossRef Medline
10. Olovnikov, A. M. (1973) A theory of marginotomy: the incomplete copying of template margin in enzymic synthesis of polynucleotides and biological significance of the phenomenon. J. Theor. Biol. 41, 181 – 190 CrossRef Medline
11. Sfeir, A., and de Lange, T. (2012) Removal of shelterin reveals the telomere end-protection problem. Science 336, 593 – 597 CrossRef Medline
12. de Bruin, D., Kantrou, S. M., Liberator, R. A., and Zakian, V. A. (2000) Telomere folding is required for the stable maintenance of telomere position effects in yeast. Mol. Cell. Biol. 20, 7991 – 8000 CrossRef Medline
13. de Lange, T. (2004) T-loops and the origin of telomeres. Nat. Rev. Mol. Cell Biol. 5, 323 – 329 CrossRef Medline
14. Griffith, J. D., Comeau, L., Rosenfield, S., Stansel, R. M., Bianchi, A., Moss, H., and de Lange, T. (1999) Mammalian telomeres end in a large duplex loop. Cell 97, 503 – 514 CrossRef Medline
15. Nakamura, T. M., and Cech, T. R. (1998) Reversing time: origin of telomerase. Cell 92, 587 – 590 CrossRef Medline
16. Belfort, M., Curcio, M. J., and Lue, N. F. (2011) Telomerase and retrotransposons: reverse transcriptases that shaped genomes. Proc. Natl. Acad. Sci. U.S.A. 108, 20304 – 20310 CrossRef Medline
17. Pardue, M. L., and DeBaryshe, P. G. (2011) Retrotransposons that maintain chromosome ends. Proc. Natl. Acad. Sci. U.S.A. 108, 20317 – 20324 CrossRef Medline
18. Tatsuke, T., Sakashita, K., Masaki, Y., Lee, J. M., Kawaguchi, Y., and Kusakabe, T. (2010) The telomere-specific non-LTR retrotransposons SART1 and TRAS1 are suppressed by Piwi subfamily proteins in the silkworm, Bombyx mori. Cell. Mol. Biol. Lett. 15, 118 – 133 CrossRef Medline
19. Silva-Sousa, R., Lopez-Panades, E., Casacuberta, E. (2012) Drosophila telomeres: an example of co-evolution with transposable elements. Genome Dyn. 7, 46 – 67 CrossRef Medline
20. Luan, D. D., Korman, M. H., Jakubczak, J. L., and Eickbush, T. H. (1993) Reverse transcription of R2B mRNA is primed by a nick at the chromosomal target site: a mechanism for non-LTR retrotransposition. Cell 72, 595 – 605 CrossRef Medline
21. Han, J. S. (2010) Non-long terminal repeat (non-LTR) retrotransposons: mechanisms, recent developments, and unanswered questions. Mob. DNA 1, 15 CrossRef Medline
22. Greider, C. W., and Blackburn, E. H. (1989) A telomeric sequence in the RNA of Tetrahymena telomerase required for telomere repeat synthesis. Nature 337, 331 – 337 CrossRef Medline
23. Maida, Y., Yasukawa, M., Furuchich, M., Lassmann, T., Possemato, R., Okamoto, N., Kasim, V., Hayashizaki, Y., Hahn, W. C., and Masutomi, K. (2009) An RNA-dependent RNA polymerase formed by TERT and the RMRP RNA. Nature 461, 230 – 235 CrossRef Medline
24. Podlevsky, I. D., and Chen, J. J.-L. (2016) Evolutionary perspectives of telomerase RNA structure and function. RNA Biol. 13, 720 – 732 CrossRef Medline
25. Bhattacharyya, A., and Blackburn, E. H. (1994) Architecture of telomerase RNA. EMBO J. 13, 5721 – 5731 CrossRef Medline
26. Webb, C. J., and Zakian, V. A. (2016) Telomerase RNA is more than a DNA template. RNA Biol. 13, 683 – 689 CrossRef Medline
27. Zappulla, D. C., and Cech, T. R. (2004) Yeast telomerase RNA: a flexible scaffold for protein subunits. Proc. Natl. Acad. Sci. U.S.A. 101, 10024 – 10029 CrossRef Medline
28. Zhang, Q., Kim, N.-K., and Feigon, J. (2011) Architecture of human telomerase RNA. Proc. Natl. Acad. Sci. U.S.A. 108, 20325 – 20332 CrossRef Medline
29. de Lange, T. (2005) Shelterin: the protein complex that shapes and safeguards human telomeres. Genes Dev. 19, 2100–2110 CrossRef Medline

30. Moser, B. A., and Nakamura, T. M. (2009) Protection and replication of telomeres in fission yeast. Biochem. Cell Biol. 87, 747–758 CrossRef Medline

31. Price, C. M., Boltz, K. A., Chaklen, M. F., Stewart, J. A., Beilstein, M. A., and Shippen, D. E. (2010) Evolution of CST function in telomere maintenance. Cell Cycle 9, 3157–3165 CrossRef Medline

32. Sun, J., Yu, E. Y., Yang, Y., Confer, L. A., Sun, S. H., Wan, K., Lee, N. F., and Lei, M. (2009) Snt1-Ten1 is a Rap2-Rap3-like complex at telomeres. Genes Dev. 23, 2900–2914 CrossRef Medline

33. Bryan, C., Rice, C., Harkisheimer, M., Schultz, D. C., and Skordalakas, E. (2013) Structure of the human telomeric Snt1-Ten1 capping complex. PLoS One 8, e66756 CrossRef Medline

34. Raffa, G. D., Raimondo, D., Sorino, C., Cugusi, S., Cenci, G., Cacchione, S., Gatti, M., and Ciapponi, L. (2010) Verrocchio, a Drosophilia OB fold-containing protein, is a component of the terminus telomere-capping complex. Genes Dev. 24, 1596–1601 CrossRef Medline

35. Biessmann, H., and Mason, J. M. (1997) Telomere maintenance without telomerase. Chromosoma 106, 63–69 CrossRef Medline

36. Kazda, A., Zellinger, B., Rössler, M., Derboven, E., Kusenda, B., and Riha, K. (2012) Chromosome end protection by blunt-ended telomeres. Genes Dev. 26, 1703–1713 CrossRef Medline

37. Wu, P., van Overbeek, M., Rooney, S., and de Lange, T. (2010) Apollo contributes to G-overhang maintenance and protects leading-end telomeres. Mol. Cell. 39, 606–617 CrossRef Medline

38. Nelson, A. D. L., and Shippen, D. E. (2012) Blunt-ended telomeres: an alternative ending to the replication and end protection stories. Genes Dev. 26, 1648–1652 CrossRef Medline

39. Du, H., Wang, Y.-B., Xie, Y., Liang, Z., Jiang, S.-J., Zhang, S.-S., Huang, Y.-B., and Tang, Y.-X. (2013) Genome-wide identification and evolutionary and expression analyses of MYB-related genes in land plants. DNA Res. 20, 437–448 CrossRef Medline

40. Horvath, M. P. (2013) Evolution of telomere binding proteins. In Madame Curie Bioscience Database [Internet]. Austin, TX: Landes Bioscience; 2000–2013. Available from: http://www.ncbi.nlm.nih.gov/books/NBK9998/

41. Theobald, D. L., Mitton-Fry, R. M., and Wuttke, D. S. (2003) Nucleic acid recognition by OB-fold proteins. Annu. Rev. Biophys. Biomol. Struct. 32, 115–133 CrossRef Medline

42. Kerr, I. D., Wadsworth, R. I. M., Cubbedu, L., Blankenfeldt, W., Naismith, J. H., and White, M. F. (2003) Insights into ssDNA recognition by the OB fold from a structural and thermodynamic study of Sulfolobus SSB protein. EMBO J. 22, 2561–2570 CrossRef Medline

43. Wong, J. M. Y., Kudrda, L., and Collins, K. (2002) Subnuclear shuttling of human telomerase induced by transformation and DNA damage. Nat. Cell Biol. 4, 731–736 CrossRef Medline

44. Masutomi, K., Possemato, R., Wong, J. M. Y., Currier, J. L., Tothova, Z., Manola, J. B., Ganesan, S., Lansdorp, P. M., Collins, K., and Hahn, W. C. (2005) The telomerase reverse transcriptase regulates chromatin state and DNA damage responses. Proc. Natl. Acad. Sci. U.S.A. 102, 8222–8227 CrossRef Medline

45. Park, J.-I., Venteicher, A. S., Hong, J. Y., Choi, J., Jun, S., Shkreli, M., Petroini, J. H. J., and de Lange, T. (2004) The telomeric protein T Reef2 binds the ATM kinase and can inhibit the ATM-dependent DNA damage response. PLoS Biol. 2, E240 CrossRef Medline

46. Karlseder, J., Hoke, K., Mizroza, O. K., Bakkenist, C., Kastan, M. B., and Petrowski, H. J., and de Lange, T. (2004) The telomeric protein T Reef2 binds the ATM kinase and can inhibit the ATM-dependent DNA damage response. PLoS Biol. 2, E240 CrossRef Medline

47. Xin, H., Liu, D., and Songyang, Z. (2008) The telosome/shelterin complex and its functions. Genome Biol. 9, 232 CrossRef Medline

48. Arnould, N., and Karlseder, J. (2015) Complex interactions between the DNA-damage response and mammalian telomeres. Nat. Struct. Mol. Biol. 22, 859–866 CrossRef Medline

49. Williams, E. S., Stoj, J., Essers, J., Ponnaiya, B., Lujisburger, M. S., Krawczyk, P. M., Ultch, R. L., Aten, J. A., and Bailey, S. M. (2007) DNA double-strand breaks are not sufficient to initiate recruitment of T Reef2. Nat. Genet. 39, 696–698; author reply 698–699 CrossRef Medline

50. Potts, R. P., and Yu, H. (2007) The SMC5/6 complex maintains telomere length in ALT cancer cells through SUMOylation of telomere-binding proteins. Nat. Struct. Mol. Biol. 14, 581–590 CrossRef Medline

51. Bian, T. M., Englelou, A., Gupta, J., Bacchetti, S., and Reddel, R. R. (1995) Telomere elongation in immortal human cells without detectable telomerase activity. EMBO J. 14, 4240–4248 CrossRef Medline

52. Shan, B., Rten, R. M., and Luizberger, M. S. (2009) Telomerase binding proteins. Nat. Struct. Mol. Biol. 17, 2347–2350 CrossRef Medline

53. Martinez, P., Thanasoula, M., Carlos, A. R., Gómez-López, G., Tjeknavorian, A. M., Schoeftner, S., Domínguez, O., Pisano, D. G., Tarsounas, M., and Blasco, M. A. (2010) Mammalian Rap1 controls telomere function and gene expression through binding to telomeric and extratelomeric sites. Nat. Cell Biol. 12, 768–780 CrossRef Medline

54. Denchi, E. L., and de Lange, T. (2007) Protection of telomeres through independent control of AT and ATR by T Reef2 and POT1. Nature 448, 1068–1071 CrossRef Medline

55. Yu, Y., Tan, R., Ren, Q., Gao, B., Sheng, Z., Zhang, J., Zheng, X., Jiang, Y., Lan, L., and Mao, Z. (2017) POT1 inhibits the efficiency but promotes the fidelity of nonhomologous end joining at non-telomeric DNA regions. Aging 9, 2529–2543 CrossRef Medline

56. Rice, C., Shastrula, P. K., Kossenkov, A. V., Bills, R., Baird, D. M., Showe, L. C., Doukou, T., Janicki, S., and Skordalakes, E. (2017) Structural and functional analysis of the human POT1-TPP1 telomeric complex. Nat. Commun. 8, 14928 CrossRef Medline

57. Bertuch, A. A., and Lundblad, V. (2003) Which end: dissecting Ku’s function at telomeres and double-strand breaks. Genes Dev. 17, 2347–2350 CrossRef Medline

58. Arbou, N., Correia, A., Ma, J., Merlo, A., Garcia-Gómez, S., Maric, M., Tognetti, M., Benny, C. W., Boulton, S. J., Saghatelian, A., and Karlseder, J. (2012) The TIN2 telomere protein T Reef associates with genomic double-strand breaks as an early response to DNA damage. Nature 477, 193–197 CrossRef Medline

59. Arnould, N., and Correia, A., Ma, J., Merlo, A., Garcia-Gómez, S., Maric, M., Tognetti, M., Benny, C. W., Boulton, S. J., Saghatelian, A., and Karlseder, J. (2012) The role of the NHEJ inhibitor CYREN. Nature 477, 548–552 CrossRef Medline
86. Nandakumar, J., Bell, C. F., Weidenfeld, I., Zaug, A. J., Leinwand, L. A., and Cech, T. R. (2012) The telomeric RNA primer mediates telomerese recruitment and processivity. *Nature* **492**, 285–289 CrossRef Medline

87. Lue, N. F. (2018) Evolving linear chromosomes and telomeres: a C-strand-centric view. *Trends Biochem. Sci.* **43**, 314–326 CrossRef Medline

88. Calvo, O., Grandin, N., Jordán-Pla, A., Mihnabes, E., González-Polo, N., Pérez-Orritín, J. E., and Charbonneau, M. (2019) The telomeric Cdc13–Stn1–Ten1 complex regulates RNA polymerase II transcription. *Nucleic Acids Res.* **47**, 6250–6268 CrossRef Medline

89. Lee, J. R., Xie, X., Yang, K., Zhang, J., Lee, Y. S., and Shippen, D. E. (2016) Dynamic interactions of *Arabidopsis* TEN1: stabilizing telomeres in response to heat stress. *Plant Cell* **28**, 2212–2224 CrossRef Medline

90. Shore, D., and Nasmuth, K. (1987) Purification and cloning of a DNA binding protein from yeast that binds to both silencer and activator elements. *Cell* **51**, 721–732 CrossRef Medline

91. Shore, D. (1994) RAIP: a protein regulator in yeast. *Trends Genet.* **10**, 408–412 CrossRef Medline

92. Teo, H., Ghosh, S., Luesch, H., Ghosh, A., Wong, E. T., Malik, N., Orbi, A., de Jesus, P., Perry, A. S., Oliver, J. D., Tran, N. L., Speiser, L. J., Wong, M., Saez, E., Schultz, P., Chanda, S. K., Verma, I. M., and Tergaonkar, V. (2010) Telomere-independent RAIP is an Ikk adaptor and regulates NF-κB-dependent gene expression. *Nat. Cell Biol.* **12**, 758–767 CrossRef Medline

93. Hussain, T., Saha, D., Purohit, G., Kar, A., Kishore Mukherjee, A., Sharma, S., Sengupta, S., Dhapola, P., Maji, B., Vedagopuram, S., Horikoshi, N. T., Horikoshi, N., Pandita, R. K., Bhattacharya, S., Bajaj, A., et al. (2017) Transcription regulation of CdkN1A (p21/CIP1/WAF1) by TRF2 is epigenetically controlled through the REST repressor complex. *Sci. Rep.* **7**, 11541 CrossRef Medline

94. Hong, J., Lee, J. H., and Chung, I. K. (2016) Telomerase activates transcription of cyclin D1 gene through an interaction with NOL1. *J. Cell Sci.* **129**, 1566–1579 CrossRef Medline

95. Ghosh, A., Saginc, G., Leow, S. C., Chatter, E., Shin, E. M., Yan, T. D., Wong, M., Zhang, Z., Li, G., Sung, W.-K., Zhou, J., Cheng, W. J., Li, S., Liu, E., and Tergaonkar, V. (2012) Telomere directly regulates NF-κB-dependent transcription. *Nat. Cell Biol.* **14**, 1270–1281 CrossRef Medline

96. Poulet, A., Pisano, S., Faivre-Moskalenko, C., Pei, B., Tauran, Y., Haefekter, Z., Brunet, F., Le Bihan, Y.-V., Ledu, M.-H., Monnet, F., Hugo, N., Amiard, S., Argoul, F., Chaboud, A., Gilson, E., and Giraud-Panis, M.-J. (2012) The N-terminal domains of TRF1 and TRF2 regulate their ability to condense telomeric DNA. *Nucleic Acids Res.* **40**, 2566–2576 CrossRef Medline

97. Dehal, P., and Boore, J. L. (2005) Two rounds of whole genome duplication in the ancestral vertebrate. *PLoS Biol.* **3*, e314 CrossRef Medline

98. Broccoli, D., Smogorzewska, A., Chong, L., and de Lange, T. (1997) Human telomeres contain two distinct Myb-related proteins, TRF1 and TRF2. *Nat. Genet.* **17**, 231–235 CrossRef Medline

99. Smogorzewska, A., van Steensel, B., Bianchi, A., Oelmann, S., Schaefer, M. R., Schnapp, G., and de Lange, T. (2000) Control of human telomere length by TRF1 and TRF2. *Mol. Cell. Biol.* **20**, 1695–1688 CrossRef Medline

100. van Steensel, B., Smogorzewska, A., and de Lange, T. (1998) TRF2 protects human telomeres from end-to-end fusions. *Cell* **92**, 401–413 CrossRef Medline

101. Lue, N. F., and Chan, J. (2013) Duplication and functional specialization of the telomere-capping protein Cdc13 in *Candida* species. *J. Biol. Chem.* **288**, 29115–29123 CrossRef Medline

102. Hockemeyer, D., Daniels, J.-P., Takai, H., and de Lange, T. (2006) Recent protection of telomeres 1 proteins POT1a and POT1b 1 Both protection of telomeres 1 proteins POT1a and POT1b can repress ATR signaling by RPA exclusion but binding to CST limits ATR repression by POT1b. *J. Biol. Chem.* **293**, 14384–14392 CrossRef Medline

103. Hagihara, M., Komatsu, T., Sugiuira, S. S., Isoda, R., Tada, H., Tanigawa, N., Kato, Y., Ishida, N., Kobayashi, K., and Matsushita, K. (2013) POT1b regulates phagocytosis and NO production by modulating activity of the small GTPase Rab5. *Biochem. Biophys. Res. Commun.* **439**, 413–417 CrossRef Medline
105. Raices, M., Verdun, R. E., Compton, S. A., Haggblom, C. I., Griffith, J. D., Dillin, A., and Karleskind, J. (2008) C. elegans telomeres contain G-strand and C-strand overhangs that are bound by distinct proteins. Cell 132, 745–757 CrossRef Medline

106. Cheng, C., Shtessel, L., Brady, M. M., and Ahmed, S. (2012) Caenorhabditis elegans POT-2 telomere represses a mode of alternative lengthening of telomeres with normal telomere lengths. Proc. Natl. Acad. Sci. U.S.A. 109, 7805–7810 CrossRef Medline

107. Shtessel, L., Lowden, M. R., Cheng, C., Simon, M., Wang, K., and Ahmed, S. (2013) Caenorhabditis elegans POT-1 and POT-2 repress telomere maintenance pathways. G3 3, 305–313 CrossRef Medline

108. Meier, B., Barber, L. I., Liu, Y., Shtessel, L., Boulton, S. J., Gartner, A., and Ahmed, S. (2009) The MRT-1 nuclease is required for DNA single strand repair and telomerase activity in vivo in Caenorhabditis elegans. EMBO J. 28, 3549–3563 CrossRef Medline

109. Wang, W., Skropp, S., Schofield, M., and Price, C. (1992) Euplotes crassus has genes encoding telomere-binding proteins and telomere-binding protein homologs. Nucleic Acids Res. 20, 6621–6629 CrossRef Medline

110. Jacob, N. K., Lescasse, R., Linger, B. R., and Price, C. M. (2007) Tetrahymanota POT1α regulates telomere length and prevents activation of a cell cycle checkpoint. Mol. Cell. Biol. 27, 1592–1601 CrossRef Medline

111. Cranert, S., Heyse, S., Linger, B. R., Lescasse, R., and Price, C. (2014) Tetrahymanota Pot2 is a developmentally regulated paralog of Pot1 that localizes to chromosome breakage sites but not to telomeres. Eukaryot. Cell 13, 1519–1529 CrossRef Medline

112. Skropp, R., Wang, W., and Price, C. (1996) rTP: a candidate telomere protein that is associated with DNA replication. Chromosoma 105, 82–91 CrossRef Medline

113. Shakirov, E. V., Perroud, P.-F., Nelson, A. D., Cannell, M. E., Quatrano, R. S., and Shippen, D. E. (2010) Protection of telomeres 1 is required for telomere integrity in the moss Physcomitrella patens. Plant Cell. 22, 1838–1848 CrossRef Medline

114. Beilstein, M. A., Renfrew, K. B., Song, X., Shakirov, E. V., Zanis, M. J., and Shippen, D. E. (2015) Evolution of the telomere-associated protein POT1α in Arabidopsis thaliana is characterized by positive selection to reinforce protein–protein interaction. Mol. Biol. Evol. 32, 1329–1341 CrossRef Medline

115. Rossignol, P., Collier, S., Bush, M., Shaw, P., and Doonan, J. H. (2007) Arabidopsis POT1A interacts with TERT-V(18), an N-terminal splicing variant of telomerase. J. Cell Sci. 120, 3678–3687 CrossRef Medline

116. Wang, F., Poddell, E. R., Zaug, A. I., Yang, Y., Baciu, P., Cech, T. R., and Lei, M. (2007) The POT1–TPP1 telomere complex is a telomerase processivity factor. Nature 445, 506–510 CrossRef Medline

117. Surovtseva, Y. V., Shakirov, E. V., Vespa, L., Osbun, N., Song, X., and Shippen, D. E. (2007) Arabidopsis POT1 associates with the telomerase RNP and is required for telomere maintenance. EMBO J. 26, 3653–3661 CrossRef Medline

118. Latchick, C. M., and Cech, T. R. (2010) POT1–TPP1 enhances telomerase processivity by slowing primer dissociation and aiding translocation. EMBO J. 29, 924–933 CrossRef Medline

119. Shakirov, E. V., McKnight, T. D., and Shippen, D. E. (2009) POT1-independent single-strand telomeric DNA binding activities in Brassicaceae. Plant J. 58, 1004–1015 CrossRef Medline

120. Arora, A., Beilstein, M. A., and Shippen, D. E. (2016) Evolution of Arabidopsis protection of telomeres 1 alters nucleic acid recognition and telomerase regulation. Nucleic Acids Res. 44, 9821–9830 CrossRef Medline

121. Shakirov, E. V., Surovtseva, Y. V., Osbun, N., and Shippen, D. E. (2005) The Arabidopsis POT1 and Pot2 proteins function in telomere length homeostasis and chromosome end protection. Mol. Cell. Biol. 25, 7725–7733 CrossRef Medline

122. Kobayashi, C. R., Castillo-González, C., Surovtseva, Y., Canal, E., Nelson, A. D. L., and Shippen, D. E. (2019) Recent emergence and extinction of the protection of telomeres 1c gene in Arabidopsis thaliana. Plant Cell Rep. 38, 1081–1097 CrossRef Medline

123. Sharma, P., Jha, A. B., Dubey, R. S., and Pessarakli, M. (2012) Reactive oxygen species, oxidative damage, and antioxidant defense mechanism in plants under stressful conditions. J. Bot. 2012, 217037 CrossRef
143. Passos, I. F., Saretzki, G., and von Zglinicki, T. (2007) DNA damage in telomeres and mitochondria during cellular senescence: is there a connection? Nucleic Acids Res. 35, 7505–7513 CrossRef Medline

144. Kovalenko, O. A., Caron, M. J., Ulema, P., Medrano, C., Thomas, A. P., Kimura, M., Bonini, M. G., Herbig, U., and Santos, J. H. (2010) A mitochondrial telomerase defective in nuclear-cytoplasmic shuttling fails to immortalize cells and is associated with mitochondrial dysfunction. Aging Cell 9, 203–219 CrossRef Medline

145. Monaghan, R. M., and Whitmarsh, A. J. (2015) Mitochondrial proteins moonlighting in the nucleus. Trends Biochem. Sci. 40, 728–735 CrossRef Medline

146. Novikova, O., and Belfort, M. (2017) Mobile group II introns as ancestral eukaryotic splicing. Trends Genet. 33, 773–783 CrossRef Medline

147. Amiard, S., Da Ines, O., Gallego, M. E., and White, C. I. (2014) Responses to telomere erosion in plants. PLoS One 9, e86220 CrossRef Medline

148. Sahin, E., Colla, S., Lisesa, M., Moslehi, J., Müller, F. L., Guo, M., Cooper, M., Kotton, D., Fabian, A. J., Walkey, C., Maser, R. S., Tonon, G., Foerster, F., Xiong, R., Wang, Y. A., et al. (2011) Telomere dysfunction induces metabolic and mitochondrial compromise. Nature 470, 359–365 CrossRef Medline

149. Webster, G., Genschel, J., Curth, U., Urbanke, C., Kang, C., and Hilgenfeld, R. (1997) A common core for binding single-stranded DNA: structural comparison of the single-stranded DNA-binding proteins (SSB) from E. coli and human mitochondria. FEBS Lett. 411, 313–316 CrossRef Medline

150. Martin, W. F., Tielens, A. G. M., Mentel, M., Garg, S. G., and Gould, S. B. (2017) The physiology of phagocytosis in the context of mitochondrial origin. Microbiol. Mol. Biol. Rev. 81, e00008–17 CrossRef Medline

151. Oh, W., Ghim, J., Lee, E.-W., Yang, M.-R., Kim, E. T., Ahn, J.-H., and Song, J. (2009) PML-IV functions as a negative regulator of telomerase by interacting with TERT. J. Cell Sci. 122, 2613–2622 CrossRef Medline

152. Minamino, T., Mitsialis, S. A., and Kourembanas, S. (2001) Hypoxia enhances the life span of vascular smooth muscle cells through telomerase activation. Mol. Cells 10, 1649–1654 CrossRef Medline

153. Minamino, T., Mitsialis, S. A., and Kourembanas, S. (2001) Hypoxia extends the life span of vascular smooth muscle cells through telomerase activation. Mol. Cell. Biol. 21, 3336–3342 CrossRef Medline

154. Yu, J., Lan, J., Wang, C., Wu, Q., Zhu, Y., Lai, X., Sun, J., Jin, C., and Huang, H. (2010) PML3 interacts with TRF1 and is essential for ALT-associated PML bodies assembly in U2OS cells. Cancer Lett. 291, 177–186 CrossRef Medline

155. Kishi, S., Wulf, G., Nakamura, M., and Lu, K. P. (2001) Telomeric protein P53 interacts with telomere-related factor PML and is down-regulated in human breast tumors. Oncogene 20, 1497–1508 CrossRef Medline

156. Kishi, S., and Gottschling, D. E. (1994) TLC1: template RNA component of Saccharomyces cerevisiae telomerase. Science 266, 404–409 CrossRef Medline

157. Shukla, S., Schmidt, J. C., Goldfarb, K. C., Cecc, T. R., and Parker, R. (2010) Inhibition of telomerase RNA decay rescues telomere length phenotype of telomere-related factors. J. Biol. Chem. 285, 1064–1072 CrossRef Medline

158. Baptista, R. J., E., Oh, B.-K., and Park, Y. N. (2009) Human PinX1 mediates TRF1 accumulation in nucleoli and enhances TRF1 binding to telomeres. J. Mol. Biol. 388, 928–940 CrossRef Medline

159. O’Connor, M. S., Safari, A., Liu, D., Qin, J., and Songyang, Z. (2004) The human Rap1 protein complex and modulation of telomere length. J. Biol. Chem. 279, 28585–28591 CrossRef Medline

160. Martin, W. F., Saretzki, G., and von Zglinicki, T. (2007) DNA damage in telomeres and mitochondria during cellular senescence: is there a connection? Nucleic Acids Res. 35, 7505–7513 CrossRef Medline

161. O’Connor, M. S., Safari, A., Liu, D., Qin, J., and Songyang, Z. (2004) The human Rap1 protein complex and modulation of telomere length. J. Biol. Chem. 279, 28585–28591 CrossRef Medline

162. Kim, S. H., Kaminker, P., and Campisi, J. (1999) TIN2, a new regulator of telomere length in human cells. Nat. Genet. 23, 405–412 CrossRef Medline

163. Ye, J. Z.-S., Hockemeyer, D., Krutchinsky, A. N., Loayza, D., Hooper, S. M., Chait, B. T., and de Lange, T. (2004) POT1-interacting protein PIP1: a telomere length regulator that recruits POT1 to the TIN2/TRF1 complex. Genes Dev. 18, 1649–1654 CrossRef Medline

164. Minamino, T., Mitsialis, S. A., and Kourembanas, S. (2001) Hypoxia extends the life span of vascular smooth muscle cells through telomerase activation. Mol. Cell. Biol. 21, 3336–3342 CrossRef Medline

165. Minamino, T., Miyachi, H., Yoshida, T., Ishida, Y., Yoshida, H., and Komuro, I. (2002) Endothelial cell senescence in human atherosclerosis: role of telomere in endothelial dysfunction. Circulation 105, 1541–1544 CrossRef Medline

166. Pendlebury, D. F., Fujiwara, Y., Tesmer, V. M., Smith, E. M., Shibuya, H., Watanabe, Y., and Nakamura, J. (2017) Dissecting the telomere–inner nuclear membrane interface formed in meiosis. Nat. Struct. Mol. Biol. 24, 1064–1072 CrossRef Medline

167. Kishi, S., Wulf, G., Nakamura, M., and Lu, K. P. (2001) Telomeric protein Pin2/TRF1 induces mitotic entry and apoptosis in cells with short telomeres and is down-regulated in human breast tumors. Oncogene 20, 1497–1508 CrossRef Medline

168. Li, B., Oestreich, S., and de Lange, T. (2000) Identification of human Rap1 protein and human telomeres. Nat. Genet. 25, 347–352 CrossRef Medline

169. Minamino, T., Mitsialis, S. A., and Kourembanas, S. (2001) Hypoxia extends the life span of vascular smooth muscle cells through telomerase activation. Mol. Cell. Biol. 21, 3336–3342 CrossRef Medline

170. Minamino, T., Miyachi, H., Yoshida, T., Ishida, Y., Yoshida, H., and Komuro, I. (2002) Endothelial cell senescence in human atherosclerosis: role of telomere in endothelial dysfunction. Circulation 105, 1541–1544 CrossRef Medline

171. Passos, I. F., Saretzki, G., and von Zglinicki, T. (2007) DNA damage in telomeres and mitochondria during cellular senescence: is there a connection? Nucleic Acids Res. 35, 7505–7513 CrossRef Medline

172. Kishi, S., Wulf, G., Nakamura, M., and Lu, K. P. (2001) Telomeric protein Pin2/TRF1 induces mitotic entry and apoptosis in cells with short telomeres and is down-regulated in human breast tumors. Oncogene 20, 1497–1508 CrossRef Medline

173. Bourns, B. D., Alexander, M. K., Smith, A. M., and Zakian, V. A. (1998) Sir proteins, Rif proteins, and Cdc13p bind Saccharomyces telomeres in vivo. Mol. Cell. Biol. 18, 5600–5608 CrossRef Medline

174. Martin, W. F., Tielens, A. G. M., Mentel, M., Garg, S. G., and Gould, S. B. (2017) The physiology of phagocytosis in the context of mitochondrial origin. Microbiol. Mol. Biol. Rev. 81, e00008–17 CrossRef Medline

175. Grozdanov, P. N., Roy, S., Kittur, N., and Meier, U. T. (2009) SHQ1 is required prior to NAF1 for assembly of H/ACA small nuclear and telomerase RNP. RNA 15, 1188–1197 CrossRef Medline

176. Teixeira, M. T., Forstemann, K., Gasser, S. M., and Lingner, J. (2002) Intracellular trafficking of yeast telomerase components. EMBO Rep. 3, 652–659 CrossRef Medline

177. Booms, B. D., Alexander, M. K., Smith, A. M., and Zakian, V. A. (1998) Sir proteins, Rif proteins, and Cdc13p bind Saccharomyces telomeres in vivo. Mol. Cell. Biol. 18, 5600–5608 CrossRef Medline