HHS Public Access

Author manuscript

DNA Repair (Amst). Author manuscript; available in PMC 2016 December 01.

Published in final edited form as:

DNA Repair (Amst). 2015 December ; 36: 137–145. doi:10.1016/j.dnarep.2015.09.017.

DNA Excision Repair at Telomeres

Pingping Jia[1], **Chengtao Her**[2], and **Weihang Chai**[1,2,*]

[1]College of Medical Sciences, Washington State University

[2]School of Molecular Biosciences, Washington State University

Abstract

DNA damage is caused by either endogenous cellular metabolic processes such as hydrolysis, oxidation, alkylation, and DNA base mismatches, or exogenous sources including ultraviolet (UV) light, ionizing radiation, and chemical agents. Damaged DNA that is not properly repaired can lead to genomic instability, driving tumorigenesis. To protect genomic stability, mammalian cells have evolved highly conserved DNA repair mechanisms to remove and repair DNA lesions. Telomeres are composed of long tandem TTAGGG repeats located at the ends of chromosomes. Maintenance of functional telomeres is critical for preventing genome instability. The telomeric sequence possesses unique features that predispose telomeres to a variety of DNA damage induced by environmental genotoxins. This review briefly describes the relevance of excision repair pathways in telomere maintenance, with the focus on base excision repair (BER), nucleotide excision repair (NER), and mismatch repair (MMR). By summarizing current knowledge on excision repair of telomere damage and outlining many unanswered questions, it is our hope to stimulate further interest in a better understanding of excision repair processes at telomeres and in how these processes contribute to telomere maintenance.

Keywords

Telomere; base excision repair; nucleotide excision repair; mismatch repair; genome stability

1. Telomere overview

Telomeres, the physical ends of linear eukaryotic chromosomes, are essential for genomic stability. In most organisms, telomeric DNA contains double-stranded G-rich tandem repeats followed by G-rich single-stranded overhangs at 3′ ends [1]. In mammalian cells, the telomeric DNA sequence is ~3–20 kb of $(TTAGGG)_n$ repeats (see section 4 on description of telomeres in yeast). The six-member telomere binding protein complex termed shelterin (TRF1, TRF2, RAP1, TIN2, POT1, and TPP1) binds to telomeric DNA, forming the unique

*Corresponding author: Weihang Chai, Ph.D., PO BOX 1495, Spokane, WA 99210, Tel: 509-358-7575, Fax: 509-358-7882, wchai@wsu.edu.

Conflict of interest statement

None.

telomeric chromatin structure called t-loop. One of the important functions of shelterin is blocking the ATM/ATR damage response pathways and suppressing double-strand break repair activities at chromosome ends, thereby preventing linear chromosome ends from erroneously being recognized as damaged DNA by the repair machinery [2, 3]. In addition, shelterin proteins control telomere length via regulating telomerase recruitment and/or modulating telomerase activity at chromosome ends (reviewed in [4]).

Telomeric DNA is vulnerable to damage in a number of ways. In addition to inevitable errors generated in the process of DNA replication during each cell division, telomeric DNA damage can also arise from oxidative stress, ionizing radiation, and carcinogenic chemicals [5–7]. Damaged telomeric DNA causes telomere dysfunction, resulting in aberrant chromosome end-to-end fusions and inappropriate recombinations that lead to genomic instability. Studies on DNA repair processes at non-telomeric chromosomal sequences have demonstrated that multiple molecular mechanisms mend damaged DNA to maintain the integrity of the genome. These mechanisms include NER, BER, MMR, non-homologous end joining (NHEJ), and homologous recombination (HR). Due to the special role of telomeres in protecting chromosome ends, DNA repair at telomeres must be tightly regulated to prevent harmful outcomes caused by unwanted repair activities. Both the NHEJ and HR pathways at telomeres have been discussed at length elsewhere [3, 8, 9]. This review focuses on NER, BER and MMR pathways, which have been loosely and sporadically studied at telomeres.

2. BER at telomeres

2.1 BER overview

BER is a cellular mechanism that corrects discrete small (non-bulky) DNA base lesions caused by oxidation, deamination, and alkylation. Defects in BER result in a higher mutation rate in a variety of organism, leading to predisposition of many types of cancer. BER is initiated by various DNA glycosylases, which recognize and remove damaged bases (see elsewhere in this issue). Eleven DNA glycosylases have been identified in humans. Both OGG1 (8-oxoguanine DNA glycosylase) and SMUG1 (single-strand-selective monofunctional uracil glycosylase 1) can remove oxidized guanine derivatives 8-oxo-7,8-dihydroguanine (8-oxo-G) and 2,6-diamino-4-hydroxy-5-formamidopyrimidine (FapyG). UNG (uracil DNA glycosylase) removes uracil, 5-hydroxyuracil, and other uracil derivatives in both single- and double-stranded DNA. AGG/MPG (alkyl guanine glycosylase/methyl purine glycosylase) removes alkylated purines and ethenopurines. TDG (thymine DNA glycosylase) removes uracil, thymine, and 5-hydroxymethyluracil. MUTYH (MutY homolog glycosylase) removes adenine when misincorporated opposite 8-oxo-G or 2,6-diamino-4-hydroxy-5-formamidopyrimidine. NTH1 (Nth Endonuclease III-Like 1), NEIL1 (Nei Endonuclease VIII-Like 1), and NEIL2 can remove oxidized pyrimidines and formamidopyrimidines.

Following the removal of the damaged base, APE1 (apurinic/apyrimidinic endonuclease 1) cuts the DNA backbone at abasic sites, leaving a single nucleotide gap. The gap is then repaired by short-patch or long-patch repair depending on the gap length. In short-patch repair, the repair gap is only one nucleotide, while in long-patch repair the gap is 2~8

nucleotides in size. In both pathways, the overhanging single-strand flap is removed by flap endonuclease 1 (FEN1). Following this incision of DNA, DNA polymerase β (Polβ) fills the gap and the single-strand break is sealed in a ligation step completed either by the DNA repair enzyme DNA ligase III in association with XRCC1 or by the replicative DNA ligase I (reviewed in [10], and see elsewhere in this issue).

2.2 BER at telomeres

The long arrays of TTAGGG repeats make telomeres particularly susceptible to oxidative lesion formation. Oxidative damages has been implicated as a primary cause of telomere shortening, evidenced by the observations that telomere attrition rate is significantly decreased when cells are grown in hypoxia condition or in the presence of antioxidant [11, 12]. In addition, both uracil residues and oxidized guanine derivatives are commonly present at telomeres [13–15]. Mounting evidence from both *in vivo* and *in vitro* studies suggests that BER is actively promoted at telomeres. Here we will review functions of each BER factor in telomere maintenance.

2.2.1 OGG1—8-oxo-G is the most abundant base lesion at telomeres. OGG1 deficiency increases oxidative guanine lesions at telomeres, leading to disruption in telomere length homeostasis [16]. Using *OGG1* knockout mice, Wang *et al*. report telomere shortening in *OGG1*−/− mouse hematopoietic cells and primary MEFs cultivated in normoxia condition (20% oxygen) or in the presence of an oxidant. Further analysis reveals other telomere abnormalities such as altered telomere sister chromatid exchanges, increased telomere single- and double-strand breaks, and preferential telomere G-strand losses in *OGG1*−/− mouse cells. These findings indicate that oxidative guanine damage affects telomere maintenance in diverse ways, and the BER pathway is required for maintaining telomere integrity in mammals [16].

The exact BER mechanism for removing telomeric 8-oxo-G residues is still unclear, and it is unknown how BER factors interplay with shelterin proteins. While the binding ability of TRF1 and TRF2 to a defined telomeric substrate is sharply reduced or abolished by 8-oxo-G incorporation [17], OGG1 incision activity is not impaired by TRF1 or TRF2, suggesting that the sequence context of telomere repeats and certain telomere configurations may be involved in telomere vulnerability due to oxidative damage [18]. Further studies are needed to elucidate BER mechanism for removing 8-oxo-G residues at telomeres.

2.2.2 UNG—Deficiency in UNG accumulates telomeric uracils and causes telomere defects including increased telomere fragility and aberrant telomere recombination in mouse hematopoietic cells [13]. Substituting thymine residues in a telomeric DNA substrate with uracils weakens POT1/TPP1 binding to substrates *in vitro* [13], indicating that telomere defects caused by UNG deficiency may be partially attributed to interference of POT1/TPP1 binding to damaged telomeric DNA.

2.2.3 NTH1—Deficiency in NTH1, another DNA glycosylase that primarily removes oxidative thymine derivative like thymine glycol, results in inefficient repair of oxidative DNA damage at telomeres, leading to preferential accumulation of oxidative base lesions at

these DNA sequences [19]. In hematopoietic tissues of *NTH1*$^{-/-}$ adult mice, telomeres suffer from multiple defects including telomere loss, increased telomere recombination, elevated telomere fragility, and DNA damage foci under normoxia. These results suggest that telomere DNA damage caused by oxidative thymine derivatives can interfere with telomere maintenance, and that NTH1 plays an important role in repairing such oxidative damage at telomeres [19].

2.2.4 NEIL—Biochemical studies show that telomeric DNA containing thymine glycol (Tg), 8-oxo-G, guanidinohydantoin (Gh), or spiroiminodihydantoin (Sp) can form quadruplex DNA structures *in vitro* [20]. The NEIL3 glycosylase has excision activity on Tg in quadruplex DNA, whereas the NEIL3 and NEIL1 glycosylases have excision activity on Sp and Gh in quadruplex DNA. These initial findings suggest that the NEIL1 and NEIL3 DNA glycosylases may remove oxidized guanine lesions from telomeres, which is crucial in telomere maintenance by preventing accumulation of oxidized guanine damages in telomeres [20, 21].

2.2.5 APE1—APE1 is the major apurinic/apyrimidinic endonuclease protecting cells from oxidative stress. It associates with telomeres and interacts with shelterin components TRF2 [22–24]. Recent report demonstrates that APE1 plays a unique role in stabilizing telomeric DNA [24]. APE1 depletion results in severe telomere defects including DNA damage response at telomeres, telomere signal loss, chromosome end fragmentation, chromosome end-to-end fusions, and extra telomeric signals regardless of cell type or telomerase status [24]. Additionally, APE1 deficiency induces telomere shortening in telomerase-expressing cells [24], consistent with reports that oxidative stress induces telomere shortening. APE1 depletion reduces TRF2 binding to telomeres [24], likely caused by the accumulation of lesions in telomeric DNA that interfere with the recruitment of factors required for telomere protection [17].

2.3 Impacts of shelterin proteins on BER activities

Nucleosomes and chromatin remodeling influence BER activity (reviewed in [25], and see elsewhere in this issue). Due to special configuration of telomere chromatin and abundant telomere binding proteins, it is likely that BER activities at telomeres are impacted by shelterin proteins. Indeed, elegant biochemical assays using purified proteins and DNA substrates demonstrate that shelterin proteins POT1, TRF1 and TRF2 physically interact with BER components Polβ, FEN1, and APE1. The shelterin proteins facilitate Polβ primer extension on actively synthesized telomeric substrates, stimulate Polα strand displacement, increase the percent of long-patch BER intermediates on telomeric substrates, as well as improve APE1 cleavage and binding [26, 27]. In addition, POT1, TRF1 and TRF2 increase FEN1 endonuclease activity on flap substrates and also increase Ligase I binding to DNA to help ligation. The stimulating effect of shelterin on BER activities may potentiate the role of telomere binding proteins in promoting efficient BER at telomeres [27].

3. NER at telomeres

3.1 NER overview

NER is an important DNA repair pathway involved in the removal of a wide spectrum of DNA lesions including UV-induced pyrimidine dimers, bulky chemical adducts, and certain forms of oxidative damage (see elsewhere in this issue). Deficiencies in various proteins participating in the NER pathway lead to three rare recessive genetic disorders: xeroderma pigmentosum (XP), Cockayne syndrome (CS), and the photosensitive form of the brittle hair disorder trichothiodystrophy (TTD). XP patients are extremely sensitive to ultraviolet rays from sunlight and are predisposed to skin cancer. CS is characterized by neurodevelopmental abnormalities, premature aging with a short lifespan (~12 years), and hypersensitivity to UV light. TTD is associated with ichthyosis and brittle hair, intellectual disability and delayed development, and recurrent infections (reviewed in [28]).

NER proteins recognize damaged sites and excise a 24–32 nt DNA fragment containing the damaged residue with extreme accuracy. Then, DNA polymerases use the undamaged strand as template to synthesize a new complementary strand to fill the gap, which is followed by ligation mediated by DNA ligase I or III [29–31]. Depending on the initial damage recognition, NER can be divided into two distinct pathways — global genome NER (GG-NER) and transcription coupled NER (TC-NER). GG-NER detects and removes lesions from the non-transcribed regions and silent chromatin over the entire genome, while TC-NER ensures faster repair of many lesions located on the sense strand of actively transcribed genes [29, 32]. GG-NER and TC-NER differ in initial damage recognition but proceed in a similar manner after recognition.

In GG-NER, DNA lesions are detected directly by the XPC/hHR23B-CETN2 complex through the structural distortion in DNA [33, 34]. This complex is the only NER factor known to be dispensable for TC-NER [35, 36]. The DDB complex, a damaged DNA-binding heterodimer consisting of DDB1 and DDB2/XPE, then recognizes DNA double-helix distortion caused by the lesion [34, 37]. Subsequently, DNA around the lesion is opened up by the concerted action of RPA, XPA and bi-directional XPB/XPG. The oligonucleotide at the lesion site is then removed, and the resulting gap is filled by DNA polymerase Polδ and Polε associated with the “sliding clamp” PCNA [34, 36].

In TC-NER, RNA polymerase II (RNAP II) recognizes a lesion while it transcribes a gene. Typically, the lesion is located in the transcribed strand, causing the RNAP II to stall or arrest [38]. When RNAP II is stalled by DNA damage, it recruits Cockayne Syndrome group B (CSB, also known as ERCC6, a member of the SNF2/SWI2 family of DNA-dependent ATPases) protein, which removes RNAP II from DNA so that it no longer acts as an obstacle to DNA repair [39, 40]. CSB, acting as a coupling factor, then recruits the CSA complex, core NER factors (XPA, TFIIH, XPG, XPF-ERCC1, and RPA), and histone acetyltransferase p300 to damaged sites. Subsequently, the promoter region is opened by the XPB and XPD helicases of TFIIH, and transcription resumes after the removal of the lesion [41].

3.2 NER at telomeres

Telomeric DNA sequence is predicted to form pyrimidine dimers after UV irradiation, and indeed, UV-induced pyrimidine dimers are detectable in the telomeric region [42, 43]. Due to the significant role of telomeres in the maintenance of chromosomal stability, NER is expected to be enhanced at telomeres. However, studies on NER at telomeres are surprisingly limited [42]. Inconsistent results have been reported on whether UV-induced pyrimidine dimers are repaired at telomeres. An early study finds that normal human somatic cells from different donor ages and disease states are capable of efficiently repairing UV-damaged telomeres, and the rate and extent of telomeric repair decline with donor's age [42]. This study also shows that in normal human fibroblasts, UV-induced pyrimidine dimers are repaired more efficiently at telomeres than those at inactive, noncoding genomic locus such as X chromosome-linked locus 754, but less efficiently than those at the actively transcribed genomic region such as the dihydrofolate reductase gene [42]. This difference may be due to the intragenomic heterogeneity of DNA repair processes that relate to the level of genomic transcription [44–48]. Supporting the conclusion that NER is proficient at telomeres, a recent finding shows that UV-induced telomeric CPDs (cyclobutane pyrimidine dimers) are repaired at a faster rate than CPDs at the bulk genome region in human skin fibroblasts expressing exogenous telomerase [49]. In striking contrast, Rochette et al [43] report that telomeres do not repair CPDs [43]. The same study shows that in two primary human fibroblast cell lines, the frequency of CPD formation at human telomeres is ~7 fold higher than that at non-telomeric region [43]. Surprisingly, cells tolerate persistent high levels of telomeric CPDs and continue proliferating without apparent acceleration in shortening of mean telomere lengths [43]. Clearly, more investigations are needed to explain the disparate results, and more importantly, to determine the NER process at telomeres.

It remains a possibility that some unrepaired lesions may be tolerated at telomeres. Tolerance of unrepaired lesions likely requires mechanisms bypassing CPDs to fully replicate telomeric DNA as well as to avoid DNA breakage and accumulation of single-stranded DNA. One such mechanism relies on specialized DNA polymerases such as Polη, which is coded by the XPV gene (this gene is defective in a variant type of xeroderma pigmentosum) and is capable of bypassing CPDs by incorporating A opposite a T or C in a CPD (reviewed in [50]). Consistently, a recent study reports that exposure to UV or hexavalent chromium (Cr(VI)) that generates bulky DNA lesions induces accumulation of Polη at telomere regions, which suppresses DNA damage foci formation at telomeres [51]. Deficiency in Polη elevates replication-associated telomere aberrations, suggesting that Polη is required for proper replication of telomeres containing bulky DNA adducts [51].

While it remains to be determined whether NER is promoted at telomeres, various NER factors have been found to play pivotal roles in telomere maintenance, implying the connection between telomere regulation and NER (see below).

3.2.1 ERCC1/XPF—ERCC1/XPF is a structure-specific endonuclease formed by ERCC1 and XPF that localizes at telomeres and interacts with TRF2 [52]. *ERCC1*−/− MEFs retain the telomeric overhang after TRF2 inhibition, suggesting that this nuclease is involved in the removal of the G-rich 3′-overhang, likely because unprotected t-loop structure resembles the

substrates for ERCC1/XPF [52]. ERCC1/XPF has also been implicated in telomere loss in mice overexpressing TRF2 [53]. Such mice show increased skin cancer after UV exposure as well as severe skin pathologies resembling human xeroderma pigmentosum with NER deficiency [53]. Keratinocytes derived from these mice are hypersensitive to UV irradiation and mitomycin C but not to ionizing radiation [53]. The TRF2 overexpressing mice have short telomeres and it appears that such telomere shortening is mediated by ERCC1/XPF [53]. Interestingly, increased TRF2 expression has been observed in human skin cancer [53]. These findings potentiate a role of TRF2 in regulating NER activity in mammalian cells.

3.2.2 XPB, XPC, XPD—Loss of XPB or XPD increases telomere attrition and enhances DNA damage at telomeres under oxidative stress, suggesting that XPB and XPD may have an important role in maintaining telomere integrity [54, 55]. Another key NER component, XPC, has been implicated in telomere maintenance as well, particularly upon UV exposure. Shorter telomeres have been observed in *XPC−/−* mice skin after chronic UV exposure, which is thought to be associated with an increased tumor incidence [56]. Interestingly, the telomere shortening effect can be reversed by an additional deficiency in telomerase. Double knockout mice deficient in both XPC and telomerase show aberrantly long telomeres that presumably results from activation of the alternative lengthening of telomeres (ALT) pathway, suggesting that XPC may have a role in suppressing aberrant telomere recombination [56]. Overall, the functions of XPB, XPC, XPD at telomeres are largely unclear.

3.2.3 CSB—CSB belongs to the SNF2/SWI2 ATPase family and is a multifunctional protein playing a role in chromatin remodeling, TC-NER, and transcription regulation (reviewed in [57]). The majority of CS patients are defective in the *CSB* gene [58, 59]. Batenburg et al. found that the CSB protein locates at a subset of telomeres and physically interacts with one shelterin component TRF2 [60], thus it is possible that CSB may be recruited to telomeres by TRF2 [60]. Deficiency in CSB affects multiple aspects of telomere homeostasis [60]. First, deficiency in CSB induces accumulation of fragile telomeres and dysfunctional telomeres [60], suggesting that CSB may be required for promoting efficient replication of telomeres and contributing to telomere protection. Consistent with this notion, treatment with aphidicolin further induces the frequency of fragile telomeres in CS cells [60], suggesting that telomere replication is compromised in CS cells. In addition, CS controls telomere length in a telomerase-dependent manner. While cells derived from CS patients undergo telomere shortening, overexpression of wild-type CSB into CS cells results in telomerase-dependent telomere lengthening [60]. The latter is associated with a reduction of the amount of telomere-bound TRF1 [60], a negative mediator of telomere length maintenance [61–63]. Furthermore, Batenburg et al. also observe that CS cells or CSB knockdown cells reduces the expression of the long non-coding telomere repeat-containing RNA known as TERRA [60]. Given that CSB regulates transcription and can function as a chromatin remodeler [57], the effect of CSB deficiency on TERRA expression is not entirely surprising. Again, current studies on CSB at telomeres are mainly descriptive, and it remains to be determined which function(s) of CSB plays a direct role in telomere maintenance.

4. BER and NER in subtelomeric and telomeric chromatin of yeast

Unlike mammalian cells, budding yeast telomeric DNA is composed of ~350 bp long $(TG_{1–3})_n$ tandem repeats that are bound by the Rap1 protein and its interacting proteins Rif1p and Rif2p [1]. These proteins negatively regulate telomere length via a protein-counting mechanism, in which the number of Rap1p-Rif1p-Rif2p bound to telomeres limits the action of telomerase in *cis*. The yeast telomerase, as its mammalian counterpart, is composed of two core components, a reverse transcriptase protein subunit (encoded by the *EST2* gene) and a template RNA (encoded by the *TLC1* gene) [64]. Unlike mammalian cells, yeast telomeres lack nucleosomes, and only the subtelomeric repeats contain nucleosomes [64]. Despite the lack of nucleosomes, telomere chromatin is repressive and forms heterochromatin. This is due to the recruitment of Sir proteins, including Sir2p, Sir3p, and Sir4p, by Rap1p and Ku70/Ku80 dimer to telomeres. Sir2p is a histone deacetylase that removes the acetyl groups from H3 and H4 histone tails, generating histone binding sites for Sir3p and Sir4p. This leads to the spreading of the Sir2-3-4 complex from sites of nucleation over a 2–3 kb subtelomeric domain. Consequently, genes near telomeres are transcriptionally silenced — a phenomenon referred to as telomere position effect (TPE) [65, 66]. The extent of silencing decreases with increasing distance from the telomere [66]. In addition, yeast telomeres gather into distinct subcompartments near the nuclear periphery [67, 68]. It is thought that clustering of telomeres helps sequestration of silencing factors to chromosome ends, therefore avoiding inappropriate repression of genes located elsewhere in the genome. However, silencing is not uniformly present at all native telomeres, with some chromosome ends exhibiting very little silencing while others having substantial silencing [69].

Like mammalian telomeres, oxidative damage occurs in telomeric guanines in *S. cerevisiae* [70]. The Liu group developed an elegant quantitative assay for measuring oxidative guanine damage at telomeres. Using this assay, they found that telomeric oxidative guanine damage can be enhanced by exogenous oxidative stress, and *ogg1*Δ cells accumulate more telomeric lesions than wild-type cells, suggesting that Ogg1 may be a primary DNA glycosylase in excising oxidized guanines in yeast telomeres [70]. Reminiscent to the effect of 8-oxo-G on TRF1 and TRF2 binding to mammalian telomeric sequences [15, 17, 70], yeast telomeric sequences containing oxidized guanine lesions reduce Rap1p and Rif2p binding to telomeres [70]. In addition, *OGG1* deletion leads to telomere lengthening that depends on telomerase and/or homologous recombination pathways, suggesting that both telomerase and recombination pathways become more active upon *OGG1* deletion [70]. Since Rap1p and Rif1/2p negatively regulates telomerase, the decreased telomeric binding of Rap1p and Rif2p explains the telomerase-dependent telomere lengthening in *ogg1*Δ cells. In addition, increased recombination rate has been observed in yeast cells with BER deficiency or harboring high density of 8-oxo-G [71, 72], and therefore, it is conceivable that Ogg1 deficiency-induced accumulation of telomeric 8-oxo-G also contributes to increased telomere recombination.

NER occurs at yeast telomeres to repair UV-induced DNA damage [73, 74], yet very little is known about how NER functions at telomeres in yeast. Histone modifications, especially acetylations, have a profound effect on NER [75, 76]. Nucleosomes with a higher level of

histone H4 acetylation stimulate repair synthesis [77]. Several reports investigated the effect of chromatin structure and histone deacetylation on NER efficiency at the subtelomeric region in budding yeast. Irizar et al. report that UV-induced CPDs are repaired significantly more efficiently at non-repressive subtelomeric ends than at repressive subtelomeric ends [74]. Such suppression of NER is modulated by Sir2p, as deletion of *SIR2* increases NER efficiency at repressed subtelomeres to a level similar to that at non-repressed subtelomeres. Consistently, Sir2p at repressive subtelomeres inhibits H3 and H4 acetylation [74]. Similarly, when telomere silencing is enhanced by either overexpression of Sir3p or deletion of *RPD3* (another histone deacetylase), NER is inhibited at subtelomeres [73]. Therefore, the NER efficiency at yeast subtelomeres appears to be controlled by the silencing state of subtelomeric chromatin structure.

5. MMR at telomeres

5.1 MMR overview

MMR is an evolutionarily conserved process involved in the recognition and correction of erroneous nucleotides including mismatched nucleotides arising during DNA replication, heteroduplexes formed during recombination, as well as DNA lesions induced by various chemical and physical factors [78–80]. Besides repairing mismatches, MMR proteins also play important roles in meiotic and mitotic recombination, DNA damage signaling, triplet-repeat expansion, as well as somatic hypermutation and class-switching recombination (reviewed in [79, 80]). Although the basic functions of MMR pathways are largely conserved from bacteria to humans, the eukaryotic MMR systems are much more complex and composed of multiple MutS and MutL homologs, which form corresponding functional heterodimeric complexes [81]. In humans, MutSα (hMSH2-MSH6) recognizes base-base mismatches and small insertion/deletion loops, whereas MutSβ (hMSH2-hMSH3) can recognize relatively larger insertion/deletion loops. The recognition of unpaired nucleotides by MutSα or MutSβ will recruit MutLα (hMLH1-hPMS2) – the major MutL heterodimer in MMR. The MutSα (or MutSβ) complex is able to search for the strand breaks in an ATP-dependent fashion and then recruits exonucleases, such as EXO1, to eliminate the DNA strand containing the incorrect nucleotide. The single-strand DNA-binding protein RPA binds to and stabilizes the single-stranded DNA, and the single-stranded gap is filled in by Polδ and ligated by Ligase I [80]. Given its biological importance, MMR deficiency frequently leads to an increased mutation rate, microsatellite instability and the development of Lynch syndrome (a hereditary colorectal cancer syndrome). The most frequently mutated MMR genes in Lynch syndrome are hMSH2 and hMLH1 [82, 83].

5.2 MMR at telomeres

5.2.1 MMR and telomere length control—A number of studies suggest that MMR deficiency can be associated with telomere shortening. Analysis of Lynch syndrome families shows that telomere lengths in leukocytes of cancer patients carrying MMR gene mutations are significantly shorter than symptom-free mutation carriers and healthy controls [84]. In addition, cancer patients with MMR gene mutations show the most pronounced telomere attrition with age [84]. However, it is entirely unknown whether the shorter telomere length in cancer patients reflects the effects of MMR deficiency or merely represents a

consequence of the cancer syndrome. Another evidence suggesting that defective MMR may impact telomere length comes from a survey of colon carcinomas [85]. The results of this study reveal that tumors with high microsatellite instability (a surrogate marker for MMR deficiency) tend to have shorter telomeres in comparison to those with stable microsatellites [85]. Moreover, down-regulation of hMSH2 in normal primary human lung fibroblasts increases the rate of telomere shortening [86]. It has been proposed that MMR deficiency may result in an accumulation of telomere mutations, thereby leading to telomeric repeat instability and accelerated telomere shortening [86].

Interestingly, results from animal model studies differ significantly from what observed in human cells. Tissues or primary MEFs derived from *MSH2*−/− mice have normal telomere lengths and telomerase activity [87]. It is highly plausible that the potential effect of MMR deficiency on telomere length differs by species and it can also be a consequence of the disease induced by MMR deficiency.

5.2.2 Role of MMR proteins in telomere DNA recombination—Results from analyzing yeast, mammalian, and human cell systems suggest that defective MMR proteins exert diverse effects on telomere DNA recombination. In budding yeast, loss of MMR proteins such as MLH1, MSH2, or PMS1 (PMS2 in mammals) promotes ALT in telomerase-deficient cells and enhances telomerase-independent cellular proliferation and survival [88]. In humans, hMSH6 deficiency in telomerase-expressing colon cancer cells induces ALT-like telomere elongation, but neither MSH2 nor MLH1 deficiency induces ALT [89]. In mammalian cells, PMS2 deficiency increases sister telomere chromatid exchange — a hallmark of ALT [90]. These initial observations suggest that a subset of but not all MMR proteins may possess MMR-independent functions in ALT, especially in the suppression of aberrant telomere recombination. It is thus very tempting to speculate that the anti-recombination function of MMR proteins may be related to such an action.

5.2.3 Role of MMR proteins in telomere damage signaling—It has been well recognized that MMR proteins also mediate DNA damage signaling. Dysfunctional telomeres are recognized as DNA damage and can activate the ATM/ATR DNA damage signaling pathways [9, 91]. Two studies investigate the role of MMR proteins in signaling dysfunctional telomeres. Using *MSH2*−/−*TERC*−/− double knockout mice, Martinez *et al.* show that abrogation of MSH2 rescues aging pathologies and tumor development associated with short telomeres [92]. Such rescue is not due to a rescue of short telomeres per se in telomerase null mice, as loss of MSH2 does not prevent telomere shortening in *TERC*−/− mice. Instead, it appears that such rescue results from MSH2 deficiency-induced attenuation of p53 and p21 induction [92], suggesting that MSH2 modulates the p53-mediated signaling pathway and cell cycle arrest in response to short and dysfunctional telomeres. This observation is consistent with the direct role of MMR proteins in signaling DNA damage through the p53 pathway [93–95].

A similar role has also been suggested for mammalian PMS2. Investigation of mice doubly deficient for telomerase and PMS2 reveals that PMS2 deficiency prolongs the lifespan and survival of telomerase-deficient mice, rescues degenerative pathologies, and reduces tumor formation [90]. Similar to the loss of MSH2, PMS2 deficiency attenuates p21 induction in

response to short telomeres [90]. Together, these results suggest that certain MMR proteins like MSH2 and PMS2 may be in the same pathway with p21 and p53, and regulate signaling of cell cycle arrest associated with telomere dysfunction. These findings highlight the important role of MMR proteins in mediating cellular response to dysfunctional telomeres *in vivo*, which is fundamental to organismal aging and to tumor development.

5.2.4 Does MMR function at telomeres?—Our current knowledge on MMR at telomeres is exceedingly limited. Many fundamental questions remain to be answered. First, are telomeres hot spots for incorporating DNA mismatches or are they more resistant to mismatch incorporation? While it is expected that the abundant Cs in telomere repeats make telomeres highly susceptible to potential U:G mismatches, telomeres form special chromatin structures and shelterin proteins may have an inhibitory effect on mismatch incorporation. In order to fully understand the telomere maintenance mechanism, it is important to apprehend whether mismatches are present at telomeres and how these mismatches are generated and recognized. Second, if mismatch incorporation is present at telomeres, to what extent do DNA mismatches affect shelterin protein binding to telomeres? To date there has been no report on the binding efficiency of shelterin proteins to DNA substrates containing mismatches. Third, are telomeres functional substrates for MMR? Does normal MMR process occur at telomeres to remove mismatches? Does the presence of shelterin proteins have an inhibitory or stimulatory effect on the MMR process? Fourth, recent research progress suggests that chromatin organization, nucleosome assembly factors, and histone modifications regulate MMR activities [96]. How does the special telomere chromatin organization, nucleosome composition, TERRA, and unique histone modifications impact the MMR process at telomeres? Lastly, it is known that MMR deficiency frequently display microsatellite instability [97, 98]. The tandem hexanucleotide telomeric repeats may conceptually be envisioned as super microsatellites. Thus, could telomere instability caused by MMR deficiency be another potential mechanism driving tumorigenesis? Answers to these questions will not only elucidate the mechanisms by which mismatches are repaired at telomeres, but also offer needed insights into the molecular basis of tumorigenesis resulting from MMR deficiency, ultimately providing guidance to design more effective therapeutic strategies.

6. Concluding remarks and future directions

The cellular and environmental processes that influence the accumulation of DNA damages at telomeres are directly relevant to cancer and aging. Although NER, BER, and MMR pathways play indispensable roles in DNA damage response and repair, excision repair at telomeres is a largely unexplored area. While it is clear that BER is active at telomeres, the roles of NER and MMR at chromosome ends are elusive. This may be partially due to the complications from either the essential functions of the core repair proteins in cell/organismal survival, or from the difficulties in separating telomere-specific roles of repair factors from their genome-wide functions. To this end, we have summarized the known functions of major BER, NER, MMR proteins at telomeres and their interacting shelterin partners in Table 1.

Several unique features of telomeres in fact warrant special considerations of excision repair. First, telomeres form the t-loop structure by inserting the single-stranded G-rich overhang back to duplex telomeric DNA [99, 100]. Formation of the t-loop structure is thought to shield chromosome ends from being recognized as double-strand breaks and help protect telomeres from being erroneously repaired. If DNA lesions are produced inside t-loop, how would these lesions be recognized and repaired by excision repair processes in the context of t-loop? Would it require temporary disassembly of t-loop? How would shelterin proteins affect BER, NER, or MMR activities at telomeres? Second, mammalian telomere chromatin contains nucleosomes [101, 102]. It is well established that efficiency of excision repair is profoundly influenced by chromatin remodeling complexes and histone modifications [96, 103–105], and DNA repair process and chromatin remodeling act cooperatively to initiate damage recognition and promote chromatin rearrangement in order to attain efficient removal of lesions [106–108]. Telomeres are specialized heterochromatin characterized by hypermethylation of DNA and repressive histone marks such as hypermethylation of histone H3 at different lysine residues, most notably at lysine 9 (H3K9), hypoacetylation of H3 and H4, and binding of heterochromatin protein 1 (HP1) to telomeres [109–111]. Additionally, both TRF1 and TRF2 are able to remodel nucleosomal spacing at telomeres [112–114]. Changes in epigenetic pattern and nucleosomal organization also occur upon deprotection of telomeres [115–118]. Is the detection of DNA damage affected by the unique telomere chromatin structure? How does the telomere chromatin structure influence the efficiency of excision repair in terms of both the access to damaged sites and the repair synthesis step? Does efficient repair require remodeling of telomere chromatin, and if so, how does such remodeling impact telomere stability? On the other hand, at least NER induces chromatin remodeling. Upon completion of NER, new H3.1 histones are incorporated at sites of newly synthesized DNA [119–121]. How does new histone incorporation impact telomere maintenance? Moreover, one recent study demonstrates that in human cells, telomeres spatially interact with specific chromosome regions and form high-order chromatin structures via chromosome looping [122]. Such chromosome looping brings the telomere close to genes up to 10 Mb away from the telomere when telomeres are long, and looping disappears when telomeres become short [122]. Therefore, telomere shortening likely alters global chromatin organization including regions distal from chromosome ends [122]. Then, does this high-order chromatin structure impact the recognition and repair of DNA lesions residing within the structure? Answers to all these questions will uncover the intricate relationship between telomere maintenance and DNA repair activities.

Lastly, given the unique role of telomeres in protecting genome stability, excision repair activities need to be tightly regulated to avoid unwanted repair activities. Although NER, BER, MMR are presumed to take place at telomeres, how these repair activities are regulated at telomeres remain entirely unknown. Emerging evidence suggests that TRF1 and/or TRF2 serve as a platform for recruiting various repair proteins to telomeres, and meanwhile, these and other shelterin proteins may play a regulatory role in restricting repair to a certain level. Further research is needed to understand the exact mechanism of excision repairs at telomeres, the consequences of excision repair deficiency on telomere stability,

and the relation of excision repair to telomere function in cancer, premature aging, and other diseases.

Acknowledgments

Work in Chai Lab is supported by NIH grant R01GM112864. The authors are grateful to P. Opresko for sharing unpublished data. We thank Ms. M. Chastain for critically reading the manuscript.

References

1. Zakian VA. Telomeres: beginning to understand the end. Science. 1995; 270:1601–1607. [PubMed: 7502069]
2. de Lange T. Shelterin: the protein complex that shapes and safeguards human telomeres. Genes Dev. 2005; 19:2100–2110. [PubMed: 16166375]
3. de Lange T. How Shelterin Solves the Telomere End-Protection Problem. Cold Spring Harb Symp Quant Biol. 2011
4. Palm W, de Lange T. How shelterin protects mammalian telomeres. Annu Rev Genet. 2008; 42:301–334. [PubMed: 18680434]
5. Cooke MS. Oxidative DNA damage: mechanisms, mutation, and disease. The FASEB Journal. 2003; 17:1195–1214. [PubMed: 12832285]
6. Iliakis G, Wang Y, Guan J, Wang H. DNA damage checkpoint control in cells exposed to ionizing radiation. Oncogene. 2003; 22:5834–5847. [PubMed: 12947390]
7. Poirier MC. Chemical-induced DNA damage and human cancer risk. Discov Med. 2012; 14:283–288. [PubMed: 23114584]
8. Doksani Y, de Lange T. The Role of Double-Strand Break Repair Pathways at Functional and Dysfunctional Telomeres. Cold Spring Harb Perspect Biol. 2014; 6
9. Longhese MP. DNA damage response at functional and dysfunctional telomeres. Genes Dev. 2008; 22:125–140. [PubMed: 18198332]
10. Wallace SS. Base excision repair: a critical player in many games. DNA repair. 2014; 19:14–26. [PubMed: 24780558]
11. von Zglinicki T. Oxidative stress shortens telomeres. Trends in biochemical sciences. 2002; 27:339–344. [PubMed: 12114022]
12. Saretzki G, Von Zglinicki T. Replicative aging, telomeres, and oxidative stress. Annals of the New York Academy of Sciences. 2002; 959:24–29. [PubMed: 11976182]
13. Vallabhaneni H, Zhou F, Maul RW, Sarkar J, Yin J, Lei M, Harrington L, Gearhart PJ, Liu Y. Defective repair of uracil causes telomere defects in mouse hematopoietic cells. The Journal of biological chemistry. 2015; 290:5502–5511. [PubMed: 25572391]
14. An N, Fleming AM, White HS, Burrows CJ. Nanopore Detection of 8-Oxoguanine in the Human Telomere Repeat Sequence. ACS nano. 2015
15. Oikawa S, Kawanishi S. Site-specific DNA damage at GGG sequence by oxidative stress may accelerate telomere shortening. FEBS letters. 1999; 453:365–368. [PubMed: 10405177]
16. Wang Z, Rhee DB, Lu J, Bohr CT, Zhou F, Vallabhaneni H, de Souza-Pinto NC, Liu Y. Characterization of oxidative guanine damage and repair in mammalian telomeres. PLoS genetics. 2010; 6:e1000951. [PubMed: 20485567]
17. Opresko PL, Fan J, Danzy S, Wilson DM 3rd, Bohr VA. Oxidative damage in telomeric DNA disrupts recognition by TRF1 and TRF2. Nucleic acids research. 2005; 33:1230–1239. [PubMed: 15731343]
18. Rhee DB, Ghosh A, Lu J, Bohr VA, Liu Y. Factors that influence telomeric oxidative base damage and repair by DNA glycosylase OGG1. DNA repair. 2011; 10:34–44. [PubMed: 20951653]
19. Vallabhaneni H, O'Callaghan N, Sidorova J, Liu Y. Defective repair of oxidative base lesions by the DNA glycosylase Nth1 associates with multiple telomere defects. PLoS genetics. 2013; 9:e1003639. [PubMed: 23874233]

20. Zhou J, Liu M, Fleming AM, Burrows CJ, Wallace SS. Neil3 and NEIL1 DNA glycosylases remove oxidative damages from quadruplex DNA and exhibit preferences for lesions in the telomeric sequence context. The Journal of biological chemistry. 2013; 288:27263–27272. [PubMed: 23926102]
21. Zhou J, Fleming AM, Averill AM, Burrows CJ, Wallace SS. The NEIL glycosylases remove oxidized guanine lesions from telomeric and promoter quadruplex DNA structures. Nucleic acids research. 2015
22. Dejardin J, Kingston RE. Purification of proteins associated with specific genomic Loci. Cell. 2009; 136:175–186. [PubMed: 19135898]
23. Lee OH, Kim H, He Q, Baek HJ, Yang D, Chen LY, Liang J, Chae HK, Safari A, Liu D, Songyang Z. Genome-wide YFP fluorescence complementation screen identifies new regulators for telomere signaling in human cells. Molecular & cellular proteomics: MCP. 2011; 10:M110 001628.
24. Madlener S, Strobel T, Vose S, Saydam O, Price BD, Demple B, Saydam N. Essential role for mammalian apurinic/apyrimidinic (AP) endonuclease Ape1/Ref-1 in telomere maintenance. Proceedings of the National Academy of Sciences of the United States of America. 2013; 110:17844–17849. [PubMed: 24127576]
25. Odell ID, Wallace SS, Pederson DS. Rules of engagement for base excision repair in chromatin. Journal of cellular physiology. 2013; 228:258–266. [PubMed: 22718094]
26. Muftuoglu M, Wong HK, Imam SZ, Wilson DM 3rd, Bohr VA, Opresko PL. Telomere repeat binding factor 2 interacts with base excision repair proteins and stimulates DNA synthesis by DNA polymerase beta. Cancer research. 2006; 66:113–124. [PubMed: 16397223]
27. Miller AS, Balakrishnan L, Buncher NA, Opresko PL, Bambara RA. Telomere proteins POT1, TRF1 and TRF2 augment long-patch base excision repair in vitro. Cell Cycle. 2012; 11:998–1007. [PubMed: 22336916]
28. McKinnon PJ. DNA repair deficiency and neurological disease. Nature reviews. Neuroscience. 2009; 10:100–112. [PubMed: 19145234]
29. Gillet LC, Scharer OD. Molecular mechanisms of mammalian global genome nucleotide excision repair. Chemical reviews. 2006; 106:253–276. [PubMed: 16464005]
30. Sugasawa K. Regulation of damage recognition in mammalian global genomic nucleotide excision repair. Mutation research. 2010; 685:29–37. [PubMed: 19682467]
31. Marteijn JA, Lans H, Vermeulen W, Hoeijmakers JH. Understanding nucleotide excision repair and its roles in cancer and ageing. Nature reviews. Molecular cell biology. 2014; 15:465–481. [PubMed: 24954209]
32. de Laat WL, Jaspers NG, Hoeijmakers JH. Molecular mechanism of nucleotide excision repair. Genes Dev. 1999; 13:768–785. [PubMed: 10197977]
33. Araki M, Masutani C, Takemura M, Uchida A, Sugasawa K, Kondoh J, Ohkuma Y, Hanaoka F. Centrosome protein centrin 2/caltractin 1 is part of the xeroderma pigmentosum group C complex that initiates global genome nucleotide excision repair. The Journal of biological chemistry. 2001; 276:18665–18672. [PubMed: 11279143]
34. Nouspikel T. DNA repair in mammalian cells: Nucleotide excision repair: variations on versatility. Cell Mol Life Sci. 2009; 66:994–1009. [PubMed: 19153657]
35. Venema J, van Hoffen A, Karcagi V, Natarajan AT, van Zeeland AA, Mullenders LH. Xeroderma pigmentosum complementation group C cells remove pyrimidine dimers selectively from the transcribed strand of active genes. Molecular and cellular biology. 1991; 11:4128–4134. [PubMed: 1649389]
36. de Boer J, Hoeijmakers JH. Nucleotide excision repair and human syndromes. Carcinogenesis. 2000; 21:453–460. [PubMed: 10688865]
37. Payne A, Chu G. Xeroderma pigmentosum group E binding factor recognizes a broad spectrum of DNA damage. Mutation research. 1994; 310:89–102. [PubMed: 7523888]
38. Spivak G, Ganesan AK. The complex choreography of transcription-coupled repair. DNA repair. 2014; 19:64–70. [PubMed: 24751236]
39. Savery NJ. The molecular mechanism of transcription-coupled DNA repair. Trends Microbiol. 2007; 15:326–333. [PubMed: 17572090]

40. Hanawalt PC, Spivak G. Transcription-coupled DNA repair: two decades of progress and surprises. Nature reviews. Molecular cell biology. 2008; 9:958–970. [PubMed: 19023283]
41. Saijo M. The role of Cockayne syndrome group A (CSA) protein in transcription-coupled nucleotide excision repair. Mechanisms of ageing and development. 2013; 134:196–201. [PubMed: 23571135]
42. Kruk PA, Rampino NJ, Bohr VA. DNA damage and repair in telomeres: relation to aging. Proceedings of the National Academy of Sciences of the United States of America. 1995; 92:258–262. [PubMed: 7816828]
43. Rochette PJ, Brash DE. Human telomeres are hypersensitive to UV-induced DNA Damage and refractory to repair. PLoS genetics. 2010; 6:e1000926. [PubMed: 20442874]
44. Evans MK, Chin KV, Gottesman MM, Bohr VA. Gene-specific DNA repair and steady state transcription of the MDR1 gene in human tumor cell lines. Oncogene. 1996; 12:651–658. [PubMed: 8637722]
45. Evans MK, Bohr VA. Gene-specific DNA repair of UV-induced cyclobutane pyrimidine dimers in some cancer-prone and premature-aging human syndromes. Mutation research. 1994; 314:221–231. [PubMed: 7513055]
46. Evans MK, Taffe BG, Harris CC, Bohr VA. DNA strand bias in the repair of the p53 gene in normal human and xeroderma pigmentosum group C fibroblasts. Cancer research. 1993; 53:5377–5381. [PubMed: 8221675]
47. Nakakura EK, McCabe SM, Zheng B, Shorthouse RA, Scheiner TM, Blank G, Jardieu PM, Morris RE. A non-lymphocyte-depleting monoclonal antibody to the adhesion molecule LFA-1 (CD11a) prevents sensitization to alloantigens and effectively prolongs the survival of heart allografts. Transplant Proc. 1993; 25:809–812. [PubMed: 8438493]
48. Evans MK, Robbins JH, Ganges MB, Tarone RE, Nairn RS, Bohr VA. Gene-specific DNA repair in xeroderma pigmentosum complementation groups A, C, D, and F. Relation to cellular survival and clinical features. The Journal of biological chemistry. 1993; 268:4839–4847. [PubMed: 8444862]
49. Parikh D, Fouquerel E, Murphy CT, Wang H, Opresko PL. Telomeres are partly shielded from UV-induced damage and proficient for photoproduct-removal by nucleotide excision repair. Nature Communications. (In press).
50. Cordonnier AM, Fuchs RP. Replication of damaged DNA: molecular defect in xeroderma pigmentosum variant cells. Mutation research. 1999; 435:111–119. [PubMed: 10556591]
51. Pope-Varsalona H, Liu FJ, Guzik L, Opresko PL. Polymerase eta suppresses telomere defects induced by DNA damaging agents. Nucleic acids research. 2014; 42:13096–13109. [PubMed: 25355508]
52. Zhu XD, Niedernhofer L, Kuster B, Mann M, Hoeijmakers JH, de Lange T. ERCC1/XPF removes the 3′ overhang from uncapped telomeres and represses formation of telomeric DNA-containing double minute chromosomes. Molecular cell. 2003; 12:1489–1498. [PubMed: 14690602]
53. Munoz P, Blanco R, Flores JM, Blasco MA. XPF nuclease-dependent telomere loss and increased DNA damage in mice overexpressing TRF2 result in premature aging and cancer. Nature genetics. 2005; 37:1063–1071. [PubMed: 16142233]
54. Gopalakrishnan K, Low GK, Ting AP, Srikanth P, Slijepcevic P, Hande MP. Hydrogen peroxide induced genomic instability in nucleotide excision repair-deficient lymphoblastoid cells. Genome Integr. 2010; 1:16. [PubMed: 21176161]
55. Ting AP, Low GK, Gopalakrishnan K, Hande MP. Telomere attrition and genomic instability in xeroderma pigmentosum type-b deficient fibroblasts under oxidative stress. J Cell Mol Med. 2010; 14:403–416. [PubMed: 19840190]
56. Stout GJ, Blasco MA. Telomere length and telomerase activity impact the UV sensitivity syndrome xeroderma pigmentosum C. Cancer research. 2013; 73:1844–1854. [PubMed: 23288511]
57. Lake RJ, Fan HY. Structure, function and regulation of CSB: a multi-talented gymnast. Mechanisms of ageing and development. 2013; 134:202–211. [PubMed: 23422418]
58. Laugel V, Dalloz C, Durand M, Sauvanaud F, Kristensen U, Vincent MC, Pasquier L, Odent S, Cormier-Daire V, Gener B, Tobias ES, Tolmie JL, Martin-Coignard D, Drouin-Garraud V, Heron

D, Journel H, Raffo E, Vigneron J, Lyonnet S, Murday V, Gubser-Mercati D, Funalot B, Brueton L, Sanchez Del Pozo J, Munoz E, Gennery AR, Salih M, Noruzinia M, Prescott K, Ramos L, Stark Z, Fieggen K, Chabrol B, Sarda P, Edery P, Bloch-Zupan A, Fawcett H, Pham D, Egly JM, Lehmann AR, Sarasin A, Dollfus H. Mutation update for the CSB/ERCC6 and CSA/ERCC8 genes involved in Cockayne syndrome. Human mutation. 2010; 31:113–126. [PubMed: 19894250]

59. Mallery DL, Tanganelli B, Colella S, Steingrimsdottir H, van Gool AJ, Troelstra C, Stefanini M, Lehmann AR. Molecular analysis of mutations in the CSB (ERCC6) gene in patients with Cockayne syndrome. American journal of human genetics. 1998; 62:77–85. [PubMed: 9443879]
60. Batenburg NL, Mitchell TR, Leach DM, Rainbow AJ, Zhu XD. Cockayne Syndrome group B protein interacts with TRF2 and regulates telomere length and stability. Nucleic acids research. 2012; 40:9661–9674. [PubMed: 22904069]
61. van Steensel B, de Lange T. Control of telomere length by the human telomeric protein TRF1. Nature. 1997; 385:740–743. [PubMed: 9034193]
62. Smogorzewska A, van Steensel B, Bianchi A, Oelmann S, Schaefer MR, Schnapp G, de Lange T. Control of human telomere length by TRF1 and TRF2. Molecular and cellular biology. 2000; 20:1659–1668. [PubMed: 10669743]
63. Ancelin K, Brunori M, Bauwens S, Koering CE, Brun C, Ricoul M, Pommier JP, Sabatier L, Gilson E. Targeting assay to study the cis functions of human telomeric proteins: evidence for inhibition of telomerase by TRF1 and for activation of telomere degradation by TRF2. Molecular and cellular biology. 2002; 22:3474–3487. [PubMed: 11971978]
64. Wright JH, Gottschling DE, Zakian VA. Saccharomyces telomeres assume a non-nucleosomal chromatin structure. Genes Dev. 1992; 6:197–210. [PubMed: 1737616]
65. Rusche LN, Kirchmaier AL, Rine J. The establishment, inheritance, and function of silenced chromatin in Saccharomyces cerevisiae. Annual review of biochemistry. 2003; 72:481–516.
66. Gottschling DE, Aparicio OM, Billington BL, Zakian VA. Position effect at S. cerevisiae telomeres: reversible repression of Pol II transcription. Cell. 1990; 63:751–762. [PubMed: 2225075]
67. Gotta M, Laroche T, Formenton A, Maillet L, Scherthan H, Gasser SM. The clustering of telomeres and colocalization with Rap1, Sir3, and Sir4 proteins in wild-type Saccharomyces cerevisiae. The Journal of cell biology. 1996; 134:1349–1363. [PubMed: 8830766]
68. Ruault M, De Meyer A, Loiodice I, Taddei A. Clustering heterochromatin: Sir3 promotes telomere clustering independently of silencing in yeast. The Journal of cell biology. 2011; 192:417–431. [PubMed: 21300849]
69. Pryde FE, Louis EJ. Limitations of silencing at native yeast telomeres. EMBO J. 1999; 18:2538–2550. [PubMed: 10228167]
70. Lu J, Liu Y. Deletion of Ogg1 DNA glycosylase results in telomere base damage and length alteration in yeast. EMBO J. 2010; 29:398–409. [PubMed: 19942858]
71. Swanson RL, Morey NJ, Doetsch PW, Jinks-Robertson S. Overlapping specificities of base excision repair, nucleotide excision repair, recombination, and translesion synthesis pathways for DNA base damage in Saccharomyces cerevisiae. Molecular and cellular biology. 1999; 19:2929–2935. [PubMed: 10082560]
72. Ohno M, Miura T, Furuichi M, Tominaga Y, Tsuchimoto D, Sakumi K, Nakabeppu Y. A genome-wide distribution of 8-oxoguanine correlates with the preferred regions for recombination and single nucleotide polymorphism in the human genome. Genome research. 2006; 16:567–575. [PubMed: 16651663]
73. Livingstone-Zatchej M, Marcionelli R, Moller K, de Pril R, Thoma F. Repair of UV lesions in silenced chromatin provides in vivo evidence for a compact chromatin structure. The Journal of biological chemistry. 2003; 278:37471–37479. [PubMed: 12882973]
74. Irizar A, Yu Y, Reed SH, Louis EJ, Waters R. Silenced yeast chromatin is maintained by Sir2 in preference to permitting histone acetylations for efficient NER. Nucleic acids research. 2010; 38:4675–4686. [PubMed: 20385597]
75. Smerdon MJ, Lan SY, Calza RE, Reeves R. Sodium butyrate stimulates DNA repair in UV-irradiated normal and xeroderma pigmentosum human fibroblasts. The Journal of biological chemistry. 1982; 257:13441–13447. [PubMed: 7142158]

76. Ramanathan B, Smerdon MJ. Changes in nuclear protein acetylation in u.v.-damaged human cells. Carcinogenesis. 1986; 7:1087–1094. [PubMed: 3087643]
77. Ramanathan B, Smerdon MJ. Enhanced DNA repair synthesis in hyperacetylated nucleosomes. The Journal of biological chemistry. 1989; 264:11026–11034. [PubMed: 2738057]
78. Bak ST, Sakellariou D, Pena-Diaz J. The dual nature of mismatch repair as antimutator and mutator: for better or for worse. Front Genet. 2014; 5:287. [PubMed: 25191341]
79. Harfe BD, Jinks-Robertson S. DNA mismatch repair and genetic instability. Annu Rev Genet. 2000; 34:359–399. [PubMed: 11092832]
80. Jiricny J. The multifaceted mismatch-repair system. Nat Rev Mol Cell Bio. 2006; 7:335–346. [PubMed: 16612326]
81. Jean M, Pelletier J, Hilpert M, Belzile F, Kunze R. Isolation and characterization of AtMLH1, a MutL homologue from Arabidopsis thaliana. Mol Gen Genet. 1999; 262:633–642. [PubMed: 10628846]
82. Peltomaki P. Lynch syndrome genes. Fam Cancer. 2005; 4:227–232. [PubMed: 16136382]
83. Peltomaki P. Deficient DNA mismatch repair: a common etiologic factor for colon cancer. Hum Mol Genet. 2001; 10:735–740. [PubMed: 11257106]
84. Segui N, Pineda M, Guino E, Borras E, Navarro M, Bellido F, Moreno V, Lazaro C, Blanco I, Capella G, Valle L. Telomere length and genetic anticipation in Lynch syndrome. PloS one. 2013; 8:e61286. [PubMed: 23637804]
85. Rampazzo E, Bertorelle R, Serra L, Terrin L, Candiotto C, Pucciarelli S, Del Bianco P, Nitti D, De Rossi A. Relationship between telomere shortening, genetic instability, and site of tumour origin in colorectal cancers. Br J Cancer. 2010; 102:1300–1305. [PubMed: 20386541]
86. Mendez-Bermudez A, Royle NJ. Deficiency in DNA mismatch repair increases the rate of telomere shortening in normal human cells. Human mutation. 2011; 32:939–946. [PubMed: 21538690]
87. Campbell MR, Wang Y, Andrew SE, Liu Y. Msh2 deficiency leads to chromosomal abnormalities, centrosome amplification, and telomere capping defect. Oncogene. 2006; 25:2531–2536. [PubMed: 16331258]
88. Rizki A, Lundblad V. Defects in mismatch repair promote telomerase-independent proliferation. Nature. 2001; 411:713–716. [PubMed: 11395777]
89. Bechter OE, Zou Y, Walker W, Wright WE, Shay JW. Telomeric recombination in mismatch repair deficient human colon cancer cells after telomerase inhibition. Cancer research. 2004; 64:3444–3451. [PubMed: 15150096]
90. Siegl-Cachedenier I, Munoz P, Flores JM, Klatt P, Blasco MA. Deficient mismatch repair improves organismal fitness and survival of mice with dysfunctional telomeres. Genes Dev. 2007; 21:2234–2247. [PubMed: 17785530]
91. Takai H, Smogorzewska A, de Lange T. DNA damage foci at dysfunctional telomeres. Curr Biol. 2003; 13:1549–1556. [PubMed: 12956959]
92. Martinez P, Siegl-Cachedenier I, Flores JM, Blasco MA. MSH2 deficiency abolishes the anticancer and pro-aging activity of short telomeres. Aging Cell. 2009; 8:2–17. [PubMed: 18986375]
93. Duckett DR, Bronstein SM, Taya Y, Modrich P. hMutSalpha- and hMutLalpha-dependent phosphorylation of p53 in response to DNA methylator damage. Proceedings of the National Academy of Sciences of the United States of America. 1999; 96:12384–12388. [PubMed: 10535931]
94. Peters AC, Young LC, Maeda T, Tron VA, Andrew SE. Mammalian DNA mismatch repair protects cells from UVB-induced DNA damage by facilitating apoptosis and p53 activation. DNA repair. 2003; 2:427–435. [PubMed: 12606123]
95. Luo Y, Lin FT, Lin WC. ATM-mediated stabilization of hMutL DNA mismatch repair proteins augments p53 activation during DNA damage. Molecular and cellular biology. 2004; 24:6430–6444. [PubMed: 15226443]
96. Li GM. New insights and challenges in mismatch repair: getting over the chromatin hurdle. DNA repair. 2014; 19:48–54. [PubMed: 24767944]

97. Imai K, Yamamoto H. Carcinogenesis and microsatellite instability: the interrelationship between genetics and epigenetics. Carcinogenesis. 2008; 29:673–680. [PubMed: 17942460]

98. Ellegren H. Microsatellites: simple sequences with complex evolution. Nature reviews. Genetics. 2004; 5:435–445.

99. Griffith JD, Comeau L, Rosenfield S, Stansel RM, Bianchi A, Moss H, de Lange T. Mammalian telomeres end in a large duplex loop. Cell. 1999; 97:503–514. [PubMed: 10338214]

100. Doksani Y, Wu JY, de Lange T, Zhuang X. Super-resolution fluorescence imaging of telomeres reveals TRF2-dependent T-loop formation. Cell. 2013; 155:345–356. [PubMed: 24120135]

101. Makarov VL, Lejnine S, Bedoyan J, Langmore JP. Nucleosomal organization of telomere-specific chromatin in rat. Cell. 1993; 73:775–787. [PubMed: 8500170]

102. Tommerup H, Dousmanis A, de Lange T. Unusual chromatin in human telomeres. Molecular and cellular biology. 1994; 14:5777–5785. [PubMed: 8065312]

103. Gong F, Kwon Y, Smerdon MJ. Nucleotide excision repair in chromatin and the right of entry. DNA repair. 2005; 4:884–896. [PubMed: 15961354]

104. Suganuma T, Workman JL. Signals and combinatorial functions of histone modifications. Annual review of biochemistry. 2011; 80:473–499.

105. Reed SH. Nucleotide excision repair in chromatin: damage removal at the drop of a HAT. DNA repair. 2011; 10:734–742. [PubMed: 21600858]

106. Gong F, Fahy D, Liu H, Wang W, Smerdon MJ. Role of the mammalian SWI/SNF chromatin remodeling complex in the cellular response to UV damage. Cell Cycle. 2008; 7:1067–1074. [PubMed: 18414052]

107. Gong F, Fahy D, Smerdon MJ. Rad4-Rad23 interaction with SWI/SNF links ATP-dependent chromatin remodeling with nucleotide excision repair. Nat Struct Mol Biol. 2006; 13:902–907. [PubMed: 17013386]

108. Zhao Q, Wang QE, Ray A, Wani G, Han C, Milum K, Wani AA. Modulation of nucleotide excision repair by mammalian SWI/SNF chromatin-remodeling complex. The Journal of biological chemistry. 2009; 284:30424–30432. [PubMed: 19740755]

109. Garcia-Cao M, O'Sullivan R, Peters AH, Jenuwein T, Blasco MA. Epigenetic regulation of telomere length in mammalian cells by the Suv39h1 and Suv39h2 histone methyltransferases. Nature genetics. 2004; 36:94–99. [PubMed: 14702045]

110. Peters AH, O'Carroll D, Scherthan H, Mechtler K, Sauer S, Schofer C, Weipoltshammer K, Pagani M, Lachner M, Kohlmaier A, Opravil S, Doyle M, Sibilia M, Jenuwein T. Loss of the Suv39h histone methyltransferases impairs mammalian heterochromatin and genome stability. Cell. 2001; 107:323–337. [PubMed: 11701123]

111. Schoeftner S, Blasco MA. A 'higher order' of telomere regulation: telomere heterochromatin and telomeric RNAs. EMBO J. 2009; 28:2323–2336. [PubMed: 19629032]

112. Galati A, Rossetti L, Pisano S, Chapman L, Rhodes D, Savino M, Cacchione S. The human telomeric protein TRF1 specifically recognizes nucleosomal binding sites and alters nucleosome structure. J Mol Biol. 2006; 360:377–385. [PubMed: 16756990]

113. Pisano S, Leoni D, Galati A, Rhodes D, Savino M, Cacchione S. The human telomeric protein hTRF1 induces telomere-specific nucleosome mobility. Nucleic acids research. 2010; 38:2247–2255. [PubMed: 20056655]

114. Galati A, Magdinier F, Colasanti V, Bauwens S, Pinte S, Ricordy R, Giraud-Panis MJ, Pusch MC, Savino M, Cacchione S, Gilson E. TRF2 controls telomeric nucleosome organization in a cell cycle phase-dependent manner. PloS one. 2012; 7:e34386. [PubMed: 22536324]

115. Galati A, Micheli E, Cacchione S. Chromatin structure in telomere dynamics. Front Oncol. 2013; 3:46. [PubMed: 23471416]

116. Doil C, Mailand N, Bekker-Jensen S, Menard P, Larsen DH, Pepperkok R, Ellenberg J, Panier S, Durocher D, Bartek J, Lukas J, Lukas C. RNF168 binds and amplifies ubiquitin conjugates on damaged chromosomes to allow accumulation of repair proteins. Cell. 2009; 136:435–446. [PubMed: 19203579]

117. Peuscher MH, Jacobs JJ. DNA-damage response and repair activities at uncapped telomeres depend on RNF8. Nat Cell Biol. 2011; 13:1139–1145. [PubMed: 21857671]

118. Rai R, Li JM, Zheng H, Lok GT, Deng Y, Huen MS, Chen J, Jin J, Chang S. The E3 ubiquitin ligase Rnf8 stabilizes Tpp1 to promote telomere end protection. Nat Struct Mol Biol. 2011; 18:1400–1407. [PubMed: 22101936]

119. Gaillard PH, Martini EM, Kaufman PD, Stillman B, Moustacchi E, Almouzni G. Chromatin assembly coupled to DNA repair: a new role for chromatin assembly factor I. Cell. 1996; 86:887–896. [PubMed: 8808624]

120. Green CM, Almouzni G. Local action of the chromatin assembly factor CAF-1 at sites of nucleotide excision repair in vivo. EMBO J. 2003; 22:5163–5174. [PubMed: 14517254]

121. Polo SE, Roche D, Almouzni G. New histone incorporation marks sites of UV repair in human cells. Cell. 2006; 127:481–493. [PubMed: 17081972]

122. Robin JD, Ludlow AT, Batten K, Magdinier F, Stadler G, Wagner KR, Shay JW, Wright WE. Telomere position effect: regulation of gene expression with progressive telomere shortening over long distances. Genes Dev. 2014; 28:2464–2476. [PubMed: 25403178]

Highlights

- Telomeres, the unique DNA-protein structures located at the ends of chromosomes, play an important role in protecting genome integrity
- Deficiency in various BER proteins accumulates oxidative lesions at telomeric sequences, disrupting telomere length homeostasis and causing telomere dysfunction
- NER factors play pivotal roles in maintaining telomere integrity, although it is unknown how the NER pathway functions at telomeres
- MMR proteins are involved in suppressing telomeric DNA recombination and modulating telomeric DNA damage signaling

Table 1

Excision repair protein functions and their interacting partners at telomeres.

Protein	Repair pathway	Effects on telomere maintenance upon deficiency	Binding partners at telomeres	References
APE1	BER	↑ telomere loss, ↑ end-to-end fusions, radial chromosomes, ↑ extra-telomeric signal.	TRF2	Madlener et al. (2013)
OGG1	BER	telomere length deregulation, ↑ T-SCE	unknown	Lu et al. (2010), Rhee et al. (2011)
UNG	BER	↑ telomere fragility, aberrant telomere recombination	unknown	Vallabhaneni et al. (2015)
NTH1	BER	↑ telomere loss, ↑ telomere recombination, ↑ telomere fragility	unknown	Vallabhaneni et al. (2013)
XPB	NER	↑ telomere attrition rate, ↑ telomere damage	unknown	Gopalakrishnan et al, (2010), Ting et al. (2010)
XPC	NER	Telomere shortening after UV exposure, activation of ALT when telomerase is deficient	unknown	Stout et al. (2013)
XPD	NER	↑ telomere attrition rate, ↑ telomere damage	unknown	Gopalakrishnan et al, (2010)
ERCC1/XPF	NER	↑ telomeric overhang afterTRF2 inhibition, ↓ telomere loss after TRF2 overexpression	TRF2	Munoz et al. (2005), Shell et al. (2008), Zhu et al. (2003)
CSB	NER	↑ fragile telomeres, ↑ telomere damage, ↓ TERRA	TRF2	Batenburg et al. (2010)
MSH2	MMR	↑ telomere attrition rate (only in human lung fibroblasts), ↑ end-to-end fusions, telomere loss, attenuates telomere damage signaling	unknown	Martinez et al. (2009), Campbell et al. (2006)
MSH6	MMR	↑ T-SCE	unknown	Bechter et al. (2004)
PMS2	MMR	↑ T-SCE, attenuates telomere damage signaling	unknown	Siegl-Cachedenier et al. (2007)

T-SCE: Telomere sister chromatid exchange.