JOURNAL OF
CONTEMPORARY MEDICINE

DOI: 10.16899/jcm.756562
J Contemp Med 2021;11(2):254-261

Review / Derleme

The effect of Telomere Lengthening on Genetic Diseases

Telomer Uzatmanın Genetik Hastalıklara Etkisi

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Abstract

Telomeres are a characteristic of chromosomes that have increasingly large significance in research. They are studied in various diseases to discover potential treatment strategies. Their most vital characteristic is their length because the length can be used to describe different characteristics about the cell, such as its age. The length of telomeres can also be used as a potential way to treat disease. This review article's purpose is to explore how telomeres can be potentially used as a method to treat genetic diseases such as trisomy 21 and cancer.

Keywords: Telomere, telomere shortening, telomere homeostasis, down syndrome, carcinoma, hepatocellular

Öz

Telomerler, araştırmalarda gittikçe artan önemi ile kromozomların bir özelliğidir. Potansiyel tedavi stratejilerini keşfetmek için çeşitli hastalıklarda incelenirler. En hayati özellikleri uzunluklarıdır çünkü uzunluk, hücre hakkında yaşı gibi farklı özellikleri tanımlamak için kullanılabilir. Telomerlerin uzunluğu, hastalığı tedavi etmenin potansiyel bir yolu olarak da kullanılabilir. Bu inceleme makalesinin amacı, telomerlerin trizomi 21 ve kanser gibi genetik hastalıkları tedavi etmek için potansiyel olarak nasıl kullanılabileceğini keşfetmektir.

Anahtar Kelimeler: Telomer, telomer kısalması, telomer homeostazı, down sendromu, karsinom, hepatoselüler

INTRODUCTION

Telomeres are the terminal ends of chromosomes that contain repetitive nucleotide sequences. They prevent chromosomal deterioration and the fusion of the chromosomes with adjacent chromosomes. Telomeres also restrict how frequently cells divide, thereby preventing malignant transformations that are caused by a buildup of mutations. Their repeated sequence of nucleotides is AGGGTT, with the complementary strand being TCCCAA, and has a TTAGGG overhang. When a cell replicates DNA, the chromosomes are shortened by approximately 25-200 bases per replication. However, telomeres are lost because of their role in protecting the chromosomes' ends, but DNA remains undamaged.

Telomere length has been studied extensively due to its potential impact on illness. One theory claims that telomeres are vital in trisomy families. Trisomy 21 is the chief abnormality of chromosomes that causes Down Syndrome. Nondisjunction errors that involve Chromosome 21 happen mostly in the oocyte, and this statistic depends primarily on the age of the mother when the sample is studied.[1] Additional studies show that Telomere loss could form classical breakage-fusion bridge cycles and dicentric chromosomes, which are then proceeded by aneuploidy and genome arrangements. Faster dementia and impairments are also caused by this, as studied in Down syndrome other many disorders that are associated with age.[2] Despite this, it is still unknown if the severity of a pre-existing cognitive impairment can predict DS cognitive deterioration rate, or if any other factors led to telomere loss and increased aging in DS patients.[3] Scientists also do not understand how advanced maternal age can be associated with the risk for DS because the exact mechanisms and triggers that are caused by telomere shortening on DS remain unknown.[4] Other factors that are considered are the metabolic profile of the mother, environmental factors, or initial telomere length at birth.[5,6]

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Received *(Geliş Tarihi)*: 22.06.2020 **Accepted *(Kabul Tarihi)*:** 31.01.2021

This review article will explore the possibility of using telomeres as a method to treat genetic diseases. Different characteristics of telomeres will be observed as to how they can be used to treat diseases. The diseases that will be discussed in this article are Trisomy 21 and different forms of cancer. This review article will summarize available research regarding telomeres and their use in treatment of diseases.

Telomere Length as a Description of Age

A telomere's length is considered to be a heritable biomarker of genomic aging.[7] When a cell divides, telomeres shorten because DNA polymerase cannot replicate the DNA strand's 3' end in its entirety.[8] Protein complexes such as the Shelterin complexes and DNA helices bind telomeres to regulate their length and structure.[9] In some cells, the enzyme telomerase, a ribonucleoprotein that has a reverse transcriptase, accessory proteins, and the RNA template TERC, maintains the telomere's length.[10] The loss of a telomere's components and telomerase has been studied extensively to observe a link between them and diseases. Premature aging is linked to shorter telomere length, as evidenced by several age-related diseases. However, it is unclear if telomere length truly played a role in these diseases or if the studied associations were caused by reverse causation. Additionally, no two individuals have the same telomere length, and telomere length is very heritable.[11,12]

Li et al. analyzed leukocyte telomere length (LTL) by pooling imputed association and densely genotyped results across large-scale European-descent studies. They used a quantitative PCR technique that could express telomere length as a telomeric repeat number (T) to a single-copy gene (S) ratio.[13,14] Any detected variant was observed to find any association with the average LTL in each cohort by way of additive models that were adjusted for cohort specific covarities, gender, and age, and the variants were then combined by way of inversevariance-weighted meta-analysis. The results indicated that, 20 sentinel variants could be associated independently genome-wide with LTL, which included six loci that were not seen with LTL. They also identified significant variants from a Singaporean Chinese population, which included the 4 loci POT1, PARP1, MPHOSPH6, and ATM. They also confirmed seven European loci (DCAF4, RTEL1, STN1, NAF1, ZNF208, and TERT). The results show that overall, genetic variation may be specific to a region, and can contribute to LTL.[7]

Li et al. also showcased shorter telomere length as more likely to cause higher age-related illnesses, they demonstrated shorter TL to other illnesses such as thyroid cancer and lymphoma. Shorter TL protected the cells from those diseases, possibly by way of limiting cell proliferation, which makes potential oncogenic mutations less common, and may stabilize DNA replication. Their findings linked nucleotide metabolism to TL regulation to gain information on the link between TL disorders that are related to age and cancer, which hints that cells that have a longer telomere length have higher dNTP levels that cause less DNA replication commitment, leading to higher proliferation and more mutations.[7]

Maternal Telomere Length on the Offspring Telomere Length

Trisomy 21 is heavily studied as a chromosome nondisjunction model. Most of the errors that nondisjunction causes happen in the oocyte, and the amount of errors on the age of the mother.[15] The errors are classified as happening during meiosis I (MI) or meiosis II (MII), which feature different mechanisms.[16] Recombinant patterns that involve telomeres increase MI risk factors, but recombinant patterns at the chromosomes' centromeres increase MII risk factors.[17-20] If maternal age truly increases errors from nondisjunction, telomeres are expected to be shorter if mothers had nondisjunction.[1]

According to Albizua et al.[21], mothers who featured a nondisjunctional error that birthed offspring with down syndrome have shorter telomere lengths than mothers who birthed offspring without Down syndrome. This suggested that the mothers of children who had DS appeared "biologically older" than the euploid children's mothers. The cases were the mothers that birthed infants who contained a nondisjunctional maternal error, which included a sample of 404 cases; the controls included the mothers that birthed an infant that had Down syndrome because of a post-zygotic mitotic error or a paternal error, which included a sample of 24 controls; mothers who had an infant without Down syndrome were also randomly drawn from the general population, in a sample of 18 mothers.[1] Over 1500 polymorphisms that were specific to chromosome 21 and spanned 21q in the probing with DS were genotyped, and Albizua et al. determined the meiotic error's parental origin by determining which parental alleles probed with trisomy 21, also enabling them to decipher whether the error occurred in meiosis I or meiosis II.[20] Parental heterozygosity in the proband at a pericentromeic marker indicated an error at meiosis I, and parental homozygosity indicated an error at meiosis II.[19-20] Linear regression models determined the maternal age T/S ratios, and other models were used to observe a correlation between meiotic outcome group and maternal age in the regression.

Albizua et al.[21] confirmed a T/S ratio that was statistically significant by using maternal age and quantitative PCR in all maternal-derived cases of nondisjunction and in controls. To do this, they analyzed their result using the cases that informed the origin stage, which included a sample of 241 cases. The cases displayed the maternal age and T/S ratio's exact and significant association, suggesting that the selected cases on the analyses that were further conducted represented the maternal errors' overall group. They then analyzed how maternal age range affects maternal telomere length, and discovered a major decrease in maternal aged T/S ratio controls and errors from meiosis I but not meiosis II. Ultimately, the results confirmed the original hypothesis, which stated younger mothers who featured a nondisjunctional error which birthed infants with Down syndrome being "biologically older" than mothers in the same age group that birthed euploid offspring.[20]

Telomeric Alternative Lengthening

Telomeric alternative lengthening and activation of telomerase are the two main mechanisms involved with the stabilization of telomere length.[22] Soft tissue sarcoma is a disease that appears to involve telomeric alternative lengthening. In cancerous cells, telomerase is activated to maintain the telomere's length. It goes about this by activating the TERT gene's promoter region, leading to the creation of the telomerase riboprotein complex's catalytic subunit.[23,24] The mutations lead to enhanced protein expression and transcription, thereby also enabling the E-twenty-six family of transcription factors to have new binding motifs.[25]

Telomeric alternative lengthening is an elongation that telomerase works on by recombination. The lengths of the telomeres can be maintained by using this technique in some cancers. For example, tumors that feature telomeric alternative lengthening display marked heterogeneity in telomere length that Southern blotting detects, and the alternative lengthening of promyelocytic leukemia bodies that are associated with telomeres.[25] The results of a study indicated that 61% of pancreatic neuroendocrine tumors exhibited telomeric alternative lengthening, causing a phenotype that perfectly correlated with the inactivation of two proteins: the death domain-associated protein 6 (DAXX) protein or the a-thalassemia/mental retardation syndrome X-linked protein (ATRX).[26,27] A dimer is then created by these proteins, displaying their importance for the telomeres' stability and incorporation of histone 3.3 into them.[28-30]

Lee et. al investigated the phenotype of telomeric alternative lengthening in liposarcoma's various subtypes and the relationship between the expression of ATRX or DAXX in liposarcoma and telomeric alternative lengthening was examined. 111 liposarcoma samples overall came from 8 patients, and the liposarcoma categories were as follows: 52 dedifferentiated liposarcomas, 28 well-differentiated liposarcomas, 11 pleomorphic sarcomas, and 20 myxoid/ round cell liposarcomas. The results revealed that the telomeric alternative lengthening was highly correlated with high grade cancer, presence of necrosis, high stage cancer, high mitotic count, and advanced age. However, these clinicopathological parameters appeared to be linked to the well-differentiated liposarcomas, the liposarcomas that were usually mitotically inactive, low grade, devoid of humor necrosis, and low stage, so they omitted the liposarcomas that were well-differentiated from the analysis, which led them to discover that telomeric alternative lengthening still had a strong association with the aforementioned factors, but found no association with the' modified FNCLCC 'grade 1 dedifferentiated liposarcoma and negative results of alternative lengthening of telomeres.[25]

Dosage of Genetic Regions' Effect on Relomere Lengthening

Scientists have generated models that can contain additional copies of regions which are synonymous to chromosome 21 to decipher how genetic segments contribute to Down syndrome phenotypes. One model in particular is the transchromosomic Tc1 mouse model that carries an extra freely-segregating copy of human chromosome 21. Since this model's inception, further analyses have combined different models to distinguish the subregions' contributions to specific DS phenotypes.[31] Its copy of Hsa21, which is nearly complete, resides in Tc1 cells and only differs in six duplications, one deletion, and 25 structural rearrangements de novo that were potentially from the model's exposure to gamma radiation during its creation.[32] The Tc1 mouse line displays phenotypes that affect the hippocampal function,[33] locomotor activities, and short term memory impairment, all of which are characteristics in a damaged Hsa21.[34,35]

Marechal et al. studied the 13 mouse genes that are between Abcg1 and U2af1, both of which are in the telomeric part of Hsa21. Mouse chromosome 17 houses The Abcg1-U2af1 region and contains 14 conserved genes: Tmprss3, *Tff1, Tff2, Tff3, Abcg1, Rsph1, Ubash3a, Pde9a, Ndufv3, Pknox1, Slc37a1, Wdr4, Cbs,* and *U2af*. In a preliminary study, the subjects that featured the Abcg1-U2af1 trisomy displayed recognition of novel objects, conserved gene overexpression, and working memory, with the exceptions of Abcg1 due to it being inactive during the genetic engineering, and U21af1, which is outside the cell.[36] In a Tc1 mouse model, all of the Abcg1-U2af1's genes are trisomic, with the Ndufv3 gene being rearranged.[37] Ms2Yah, which is the monosomy that corresponds to the Abcg1-U2af1 region, lacks the 12 genes but carries the last exons of Abcg1, and showed social recognition defects and fear conditioning.[38] In their main study, the Ms2Yah was paired with the Tc1 model to observe how different behavioral phenotypes are affected.[31]

The results indicated that the Tc1/Ms2Yah mice and the controls learned the platform location equally quickly, but the Tc1 learned it more slowly. Learning memory was unaffected, so Marechal et al. suspect that the Tc1 mice's worse performance was due to an absence of cognitive flexibility. This decreased flexibility reinforced the result that Abcg1-U2af1's increase will affect cognitive thinking, which is also observed in Down syndrome patients.[39] Tc1 mice also exhibited major deficits in motor skills, which was tested when the mice learned to stay on a rotarod. In the learning phase, Tc1/Ms2Yah mice and Tc1 mice's performances were unimproved. This demonstrated the alteration of locomotor function's learning mechanisms, which could not be resolved by the Abcg1-U2af1's decreasing number of copies.[31]

DNA Repair's Effects On Telomeres

DNA damage is caused by exogenous factors or endogenous sources. Any damaged DNA can lead to genomic instability if left untreated, and may eventually grow tumors. Telomeres prevent these genomic instabilities from happening. Base excision repair, nucleotide excision repair, and mismatch repair are the three mechanisms of DNA repair.

1. Base Excision Repair's Effects On Telomeres

Base excision repair fixes lesions of small DNA bases that are caused by deamination, alkylation and oxidation. The lack of BER leads to a higher mutation rate, and can lead to many forms of cancer. DNA glycosylases remove the damaged bases to start BER. After the damaged bases are removed, Apyrimidinic/apurinic endonuclease 1 (APE1) slices the backbone of the DNA at apurinic or apyrimidinic sites to leave behind a single nucleotide gap, followed by a long-patch repair or a short-patch repair.[40]

In base excision repair, telomeres are susceptible to oxidative lesion formation because of their long TTAGGG repeats. Oxidative damages can reinforce shortening of the telomere, which is shown by telomere attrition rate significantly decreasing when cells mature in hypoxic conditions or with an antioxidant near them.[41,42] Telomeres feature oxidized guanine derivatives and uracil.[43–45] There is increasingly large evidence that both in vitro and in vivo studies actively promote telomeric BER.[40]

Telomeric oxidative guanine lesions are caused by a deficiency in OGG1, thereby disrupting telomere length homeostasis. In an experiment conducted by Wang et al., primary MEFs and OGG1−/− mouse hematopoietic cells had shortened telomeres with normal oxygen concentration (20%) or when an oxidant was present. Other telomeric abnormalities which were featured in OGG1−/− mouse cells included lost preferential telomere G-strands, altered telomere sister chromatid exchanges, and a higher presence of telomere singleand doublestrand breaks. These results confirm the the BER pathway's cruciality in maintaining telomeric integrity in mammals.[46] Telomeric 8-oxo-G residues removal by way of BER remains unknown along with the role BER has with shelterin. 8-oxo-G incorporation abolishes or majorly reduces TRF1 and TRF2's ability to bind to a certain telomeric substrate,[47] but TRF1 and TRF2 have no effect on OGG1 incision activity. These data imply that certain telomere configurations and the sequence context of telomere repeats cause oxidative damage to the telomere, thereby weakening it.[48]

2. Nucleotide Excision Repair's Effects On Telomeres

Nucleotide excision repair repairs DNA by removing lesions such certain forms of oxidative damage, bulky chemical adducts, and UV-induced pyrimidine dimers.[49]

NER proteins excise a fragment that is 24-32 nt that contains the damaged residue. DNA polymerases fill the gap by synthesizing a new complementary strand from the undamaged strand, and DNA ligase I or III ligates the two strands together to complete this process.[50-52] NER undertakes two distinct pathways which depend on the recognition of initial damage: global genome nucleotide excision repair (GG-NER), which erases and detects lesions from the silent chromatin and any gene that is not transcribed throughout the whole genome; and transcription coupled nucleotide excision repair (TC-NER), which repairs the sense strand's lesions more quickly.[50,53] These pathways proceed in an identical manner after initial damage recognition.[40]

Telomeric DNA sequences are hypothesized to create pyrimidine dimers following Ultraviolet irradiation, and these dimers can be detected at the telomere.[54,55] Scientists are expecting NER to be enhanced at telomeres because of the importance of telomeres in maintaining the chromosome's stability. NER studies regarding telomeres are currently uncommon,[54] and led to the reports of inconsistent results on the dimers being potentially fixed.[50] One study shows that non-mutated somatic cells that are in different states of disease and donor ages can effectively repair telomeres that were damaged by UV light, and as the donor's age increases, the extent and rate of telomeric repair decreases.[54] According to a finding, NER repairs Ultraviolet-induced cyclobutane pyrimidine dimers (CPDs) that are located on telomeres faster than CPDs that are on the human skin fibroblasts' bulk region that express exogenous telomerase, which supports that NER can function well at telomeres.[56] Rochette et al.,[55] in contrast, dismiss CPD repair of NER,[54] but confirmed CPD formation is 7 times higher at telomeres than at the regions without telomeres.[55] More studies must be done to determine how NER works at telomeres.

Unrepaired lesion tolerance potentially requires mechanisms that can bypass CPDs to completely replicate the telomeric DNA and avoid breaking apart DNA and accumulating singlestranded DNA. One of these mechanisms involves specialized DNA polymerases such as Polη, the polymerase that the XPV gene (which does not function in a variant type of xeroderma pigmentosum) codes for and can bypass CPDs by incorporating adenine to a thymine or cytosine in a CPD.[57] Consistently, exposure to hexavalent chromium (Cr(VI)) or ultraviolet light that creates sturdy DNA lesions will enable Polη to accumulate at telomeres, which suppresses the formation of telomeric DNA damage foci. Polη deficiency heightens telomeric aberrations that are associated with replication, which suggests Polη's necessity to properly replicate the telomeres if they contain bulky DNA adducts.[58] Scientists currently cannot conclude if telomeres can promote NER, but numerous NER factors are confirmed to have vital jobs in the maintenance of telomere, which implies the potential connection between NER and telomere regulation.[40]

3. Mismatch Repair's Effects On Telomeres

Mismatch repair recognizes and corrects nucleotides in erroneous positions, which include mismatched nucleotides that are a side-effect of DNA replication, heteroduplexes that are formed during recombination, and chemically or physically-induced DNA lesions.[59-61] MMR proteins also act during mitotic and meiotic recombination, triplet-repeat expansion, DNA damage signaling, class-switching recombination, and somatic hypermutation.[60,61] In eukaryotes, functional heterodimeric complexes are formed by multiple MuSt and MutL homologs, which make up MMR systems.[62] MutSαα (hMSH2-MSH6) recognizes base-base mismatches and small insertion/deletion loops, whereas MutSβ (hMSH2-hMSH3) recognizes large insertion/deletion loops. RPA stabilizes the

single-stranded DNA, and Polδ fills the stranded gap and stabilizes, which is then ligated by Ligase I.[61] MMR deficiency commonly causes micro-satellite instability, Lynch syndrome development, and an increased rate of mutation. Lynch syndrome most commonly mutates the hMSH2 and hMLH1 genes.[63,64]

MMR deficiency is hypothesized to be linked to shortening of telomeres. An analysis that studied families that had members that featured Lynch syndrome displays that telomere lengths that contained MMR gene mutations were significantly shorter in cancer patients' leukocytes than healthy controls and mutation carriers who did not exhibit symptoms. Additionally, as age increased, attrition of telomeres increased in MMR gene-mutated patients.[65] Despite these results, the scientists could not conclude whether the cancer patients' shorter telomere length truly reflects MMR deficiency's effect or only represents one of the cancer's consequences. According to a study that involves colon carcinomas, defective MMR may have an effect on telomere length, as demonstrated by the results, which state that tumors that have a high microsatellite instability (which can deduce MMR deficiency) have shorter telomere lengths in comparison to the tumors that feature stable microsatellites.[66] An additional finding of telomere shortening is that telomeres shorten as hMSH2 in normal lung fibroblasts are down regualted. MMR deficiency may lead to accumulating mutations in telomeres, which then cause accelerated telomere shortening and telomeric repeat instability.[67] Interestingly, animal model studies show significantly different results from human cell studies, showing how MMR deficiency may affect telomere length differently depending on the species. This is confirmed by primary MEFs or tissues deriving from MSH2−/− mice having normal telomerase activity and lengths of telomeres.[68]

It is currently unknown if MMR actually functions on telomeres. For example, it is unknown if telomeres can incorporate or are resistant to DNA mismatches. While the abundance of cytosines in the telomere repeats theoretically makes telomeres vulnerable to mismatches with uracil to guanine, telomeres create special chromatin structures whose shelterin proteins can inhibit mismatch incorporation. If scientists are to fully comprehend the mechanism of telomere maintenance, they must be able to conclude if mismatches can actually occur at telomeres and if so, how the mismatches are created and interpreted.[40] How mismatch incorporation affects the efficiency of the telomere's shelterin proteins on the DNA mismatch is also unknown. New research suggests that assembly factors of nucleosomes, modifications of histones, and chromatin organization regulate the activities of MMR.[69] However, MMR deficiencies are confirmed to frequently display instability of microsatellites.[70,71]

Effect of Cancer on Telomeres

Cancer is often defined as rapid cell division. While the overall mechanism of cancer's effect on telomeres remains unknown, scientists are studying possible effects.

1. Childhood cancer survivors' Telomere Attrition

Song et. al measured the length of telomeres in the leukocytes of participants who lacked cancer and explored how LTL is associated with different treatments of cancer, variation in individual health behavior, and diagnosis of comorbid health conditions.[72] Their results showed that the childhood cancer survivors exhibited significantly shorter LTL within all cancer subgroupings (which included tumors of the central nervous system, Hodgkin lymphoma, sarcomas, neuroblastoma, non-Hodgkin lymphoma, acute lymphoblastic leukemia, and Wilm's tumor) following adjustments for sex, polymorphisms that were associated with LTL, ancestry, and age when DNA sampling took place. The average LTL value was 36.8 years of age for survivors and for controls, it was 48.2 years of age. These data suggest a telomere attrition that was accelerated by approximately eleven years among survivors of cancer from childhood. In survivors, as age increased, LTL decreased at a similar rate to the controls, which indicated that many telomere reserves were lost after treatment, but their loss was not accelerated.[72-73]

Song et al. also studied LTL for potentially being associated with overall mortality and various chronic health conditions. Shorter LTL had a correlation with more common diagnosis of 14 chronic conditions, which included: cardiomyopathy, chronic hepatitis C, cholecystitis, hypercholesterolemia, hypertriglyceridemia, gastritis/duodenitis, fibrosis/cirrhosis, gastrointestinal ulcer, hypertension, headaches, obesity, obstructive and restrictive pulmonary deficits, and lymphatic infections. Higher overall mortality and shorter LTL were revealed to have a potential association, but it was statistically insignificant (P=0.08).[72] Longer LTL showed a tremendously greater chance of causing secondary thyroid cancers, and the finding supports that longer LTL can cause numerous cancers, including cancers that occur in childhood. [74] Some of the health conditions that were analyzed were potentially diagnosed before or after DNA sampling took place. These separate analyses revealed that restrictive pulmonary deficit risk and shorter LTL were related, along with hypertriglyceridemia and obstructive pulmonary deficit also being potentially related. This implies that LTL can affect childhood cancer survivors prognostically.[72-73]

One vital finding in this study was that factors in behavior or lifestyle can modify childhood cancer survivors' telomere attrition rates. Diet, physical activity, alcohol overconsumption, tobacco use, and resistance training enabled Song et al. to create a composite score which revealed that among survivors aged 18-35 years old, significantly longer age-adjusted LTL was caused by favorable health behaviors. On the contrary, the score also revealed that ageadjusted LTL among survivors that were older than 35 years of age was similar regardless of health behaviors. The result potentially hints about a critical discovery that follows childhood cancer treatment, which is that any healthy lifestyle modification can heavily impact telomere

attrition rates. The result also reinforces the substantial loss of telomere reserves that seemed to happen after treatment, which led to similar attrition rates among survivors and controls that lack cancer.[72-73]

2. Using HKR3 to Inhibit hTERT in Hepatocellular Carcinoma Cells to Regulate the Cell Cycle

Hepatocellular carcinoma is a very common form of cancer.[75] Its progression is characterized by genetic and epigenetic abnormalities that eventually enable the cancerous cells to proliferate and escape apoptosis.[76] Scientists are studying how HCC cells regulate apoptosis and the cell cycle, which can lead to better clinical management.[77] The enzyme telomerase confers immortality to cancer cells by synthesizing telomeric repeat at the chromosomes' ends and replacing the lost end sequences with new sequences during each cell division, granting the cell a longer life cycle and enabling it to divide indefinitely. A kind of telomerase called human telomere transferase (hTERT), which is expressed as a fetus develops and is eventually deactivated in adult tissues, activates HCC. hTERT's mechanism in the regulation of changes of expression during HCC is currently unknown, leading to the importance of deciphering how to regulate apoptosis and the cell cycle.[78]

In cancer cells, hTERT is over-expressed to create telomeres as a mechanism to prevent apoptosis.[11,79] However, it is still unknown if hTERT inhibits other factors to reinforce the cell cycle. Choi et al. hypothesized that if hTERT is somehow regulated, an anti-tumor strategy could be developed in HCC management by using human kruppel-related 3 and factors that are involved in the cell cycle of HCC cell lines. According to their results, hTERT was approximately 15 times more expressed in HCC tissues expression than in normal tissues when HKR3 is uninvolved. When they inhibited hTERT, they observed changes in genes relating to apoptosis, cell cycles, and senescence, which enabled them to confirm that HKR3 and hTERT had a correlation.[78]

Additional findings stated that HKR3 was hardly expressed within HCC patients' cancer tissues, but was more expressed around the liver tissue's bad prognosis, and hTERT was more prevalent in the HCC cell strains. When they found that hTERT expression and HKR3 expression were inversely related, they also confirmed that CDKNN2A expression was increased, which reduced the cyclin A1, B1, and D1 expressions. Choi et al. confirmed that hTERT inhibition causes apoptosis, so they investigated how the cell cycle was controlled in HCC cells. This was reinforced by the finding that the majority of the genetic changes were in apoptosis genes. Overall, the results of their experiment concluded that using HKR3 to inhibit hTERT can prevent the progress of HCC.[78]

Future Directions/Conclusion

The experiments that were observed in this review article are highly beneficial to our understanding of using telomeres to treat diseases. Our current understanding is that while these studies are limited, adjusting the lengths of telomeres are useful in treating diseases such as trisomy 21. However, these experiments were not without their limitations. For example, while it is confirmed that the increase maternal age leads to a greater risk of birthing a DSinfected child, the exact mechanism, triggers, and association remain unknown.[4] If a mechanism is discovered, scientists can test treatments for it, which is a solid step for successful treatment of Down Syndrome.

Consequently, it can be difficult to draw conclusions of studies with very limited data and research on telomeres' effects on diseases currently available. To achieve more precise results, the mechanisms of telomeres' effect on diseases must be studied more thoroughly. One example of this is MMR's true function on telomeres being unknown. The effect of DNA mismatch incorporation on telomeres is currently unknown, but scientists hypothesize that telomeres that can form shelterin proteins and special chromatin structures potentially inhibiting mismatch incorporation. If scientists can eventually apprehend the possibility of mismatches appearing at telomeres and how the mismatches can be recognized and generated, this will lead to a greater understanding on MMR's function on telomeres, which can lead to further research to study how telomeres can be used to treat diseases.

ETHICAL DECLARATIONS

Status of Peer-review: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: This study was supported by Barry University Biomedical Sciences Program, Miami Shores, FL 33161 USA.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Albizua I, Rambo-Martin BL, Allen EG, He W, Amin AS, Sherman SL. Association between telomere length and chromosome 21 nondisjunction in the oocyte. Hum Genet. 2015;134(11-12):1263-70.
2. Honig LS, Kang MS, Schupf N, Lee JH, Mayeux R. Association of shorter leukocyte telomere repeat length with dementia and mortality. Arch Neurol. 2012;69(10):1332-9.
3. Temple V, Jozsvai E, Konstantareas MM, Hewitt TA. Alzheimer dementia in Down's syndrome: the relevance of cognitive ability. J Intellect Disabil Res. 2001;45(Pt 1):47-55.
4. Gruszecka A, Kopczyński P, Cudziło D, et al. Telomere shortening in Down syndrome patients--when does it start?. DNA Cell Biol. 2015;34(6):412-7.
5. Révész D, Milaneschi Y, Verhoeven JE, Penninx BW. Telomere length as a marker of cellular aging is associated with prevalence and progression of metabolic syndrome. J Clin Endocrinol Metab. 2014;99(12):4607-15.
6. Nakamura K, Ishikawa N, Izumiyama N, et al. Telomere lengths at birth in trisomies 18 and 21 measured by Q-FISH. Gene. 2014;533(1):199-207.
7. Li C, Stoma S, Lotta LA, et al. Genome-wide Association Analysis in Humans Links Nucleotide Metabolism to Leukocyte Telomere Length. Am J Hum Genet. 2020;106(3):389-404.

8. Allsopp RC, Vaziri H, Patterson C, et al. Telomere length predicts replicative capacity of human fibroblasts. Proc Natl Acad Sci U S A. 1992;89(21):10114-8.
9. de Lange T. Shelterin-Mediated Telomere Protection. Annu Rev Genet. 2018;52:223-47.
10. Blackburn EH, Collins K. Telomerase: an RNP enzyme synthesizes DNA. Cold Spring Harb Perspect Biol. 2011;3(5):a003558.
11. Njajou OT, Cawthon RM, Damcott CM, et al. Telomere length is paternally inherited and is associated with parental lifespan. Proc Natl Acad Sci U S A. 2007;104(29):12135-9.
12. Broer L, Codd V, Nyholt DR, et al. Meta-analysis of telomere length in 19,713 subjects reveals high heritability, stronger maternal inheritance and a paternal age effect. Eur J Hum Genet. 2013;21(10):1163-8.
13. Cawthon RM. Telomere measurement by quantitative PCR. Nucleic Acids Res. 2002;30(10):e47.
14. Cawthon RM. Telomere length measurement by a novel monochrome multiplex quantitative PCR method. Nucleic Acids Res. 2009;37(3):e21.
15. Freeman SB, Allen EG, Oxford-Wright CL, et al. The National Down Syndrome Project: design and implementation. Public Health Rep. 2007;122(1):62-72.
16. Sherman SL, Allen EG, Bean LH, Freeman SB. Epidemiology of Down syndrome. Ment Retard Dev Disabil Res Rev. 2007;13(3):221-7.
17. Lamb NE, Freeman SB, Savage-Austin A, et al. Susceptible chiasmate configurations of chromosome 21 predispose to non-disjunction in both maternal meiosis I and meiosis II. Nat Genet. 1996;14(4):400-5.
18. Lamb NE, Sherman SL, Hassold TJ. Effect of meiotic recombination on the production of aneuploid gametes in humans. Cytogenet Genome Res. 2005;111(3-4):250-5.
19. Oliver TR, Feingold E, Yu K, et al. New insights into human nondisjunction of chromosome 21 in oocytes. PLoS Genet. 2008;4(3):e1000033.
20. Oliver TR, Tinker SW, Allen EG, et al. Altered patterns of multiple recombinant events are associated with nondisjunction of chromosome 21. Hum Genet. 2012;131(7):1039-46.
21. Warburton D. Biological aging and the etiology of aneuploidy. Cytogenet Genome Res. 2005;111(3-4):266-72.
22. Goldberg AD, Banaszynski LA, Noh KM, et al. Distinct factors control histone variant H3.3 localization at specific genomic regions. Cell. 2010;140(5):678-91.
23. Horn S, Figl A, Rachakonda PS, et al. TERT promoter mutations in familial and sporadic melanoma. Science. 2013;339(6122):959-61.
24. Huang FW, Hodis E, Xu MJ, Kryukov GV, Chin L, Garraway LA. Highly recurrent TERT promoter mutations in human melanoma. Science. 2013;339(6122):957-9.
25. Lee JC, Jeng YM, Liau JY, Tsai JH, Hsu HH, Yang CY. Alternative lengthening of telomeres and loss of ATRX are frequent events in pleomorphic and dedifferentiated liposarcomas. Mod Pathol. 2015;28(8):1064-73.
26. Jiao Y, Shi C, Edil BH, et al. DAXX/ATRX, MEN1, and mTOR pathway genes are frequently altered in pancreatic neuroendocrine tumors. Science. 2011;331(6021):1199-203.
27. Heaphy CM, de Wilde RF, Jiao Y, et al. Altered telomeres in tumors with ATRX and DAXX mutations. Science. 2011;333(6041):425.
28. Drane P, Ouararhni K, Depaux A et al. The death-associated protein DAXX is a novel histone chaperone involved in the replication-independent deposition of H3.3. Genes Dev 2010;24:1253–65.
29. Goldberg AD, Banaszynski LA, Noh KM et al. Distinct factors control histone variant H3.3 localization at specific genomic regions. Cell 2010;140:678–691.
30. Wong LH, McGhie JD, Sim M et al. ATRX interacts with H3.3 in maintaining telomere structural integrity in pluripotent embryonic stem cells. Genome Res 2010;20:351–60.
31. Marechal D, Lopes Pereira P, Duchon A, Herault Y. Dosage of the Abcg1-U2af1 region modifies locomotor and cognitive deficits observed in the Tc1 mouse model of Down syndrome. PLoS One. 2015;10(2):e0115302.
32. Gribble SM, Wiseman FK, Clayton S, et al. Massively parallel sequencing reveals the complex structure of an irradiated human chromosome on a mouse background in the Tc1 model of Down syndrome. PLoS One. 2013;8(4):e60482.
33. O'Doherty A, Ruf S, Mulligan C, et al. An aneuploid mouse strain carrying human chromosome 21 with Down syndrome phenotypes. Science. 2005;309(5743):2033-7.
34. Galante M, Jani H, Vanes L, et al. Impairments in motor coordination without major changes in cerebellar plasticity in the Tc1 mouse model of Down syndrome. Hum Mol Genet. 2009;18(8):1449-63.
35. Morice E, Andreae LC, Cooke SF, et al. Preservation of long-term memory and synaptic plasticity despite short-term impairments in the Tc1 mouse model of Down syndrome. Learn Mem. 2008;15(7):492-500.
36. Pereira PL, Magnol L, Sahún I, et al. A new mouse model for the trisomy of the Abcg1-U2af1 region reveals the complexity of the combinatorial genetic code of down syndrome. Hum Mol Genet. 2009;18(24):4756-69.
37. Gribble SM, Wiseman FK, Clayton S, et al. Massively parallel sequencing reveals the complex structure of an irradiated human chromosome on a mouse background in the Tc1 model of Down syndrome. PLoS One. 2013;8(4):e60482.
38. Sahún I, Marechal D, Pereira PL, et al. Cognition and hippocampal plasticity in the mouse is altered by monosomy of a genomic region implicated in Down syndrome. Genetics. 2014;197(3):899-912.
39. Rustay NR, Wahlsten D, Crabbe JC. Influence of task parameters on rotarod performance and sensitivity to ethanol in mice. Behav Brain Res. 2003;141(2):237-49.
40. Jia P, Her C, Chai W. DNA excision repair at telomeres. DNA Repair (Amst). 2015;36:137-45.
41. von Zglinicki T. Oxidative stress shortens telomeres. Trends Biochem Sci. 2002;27(7):339-44.
42. Saretzki G, Von Zglinicki T. Replicative aging, telomeres, and oxidative stress. Ann N Y Acad Sci. 2002;959:24-9.
43. Vallabhaneni H, Zhou F, Maul RW, et al. Defective repair of uracil causes telomere defects in mouse hematopoietic cells. J Biol Chem. 2015;290(9):5502-11.
44. An N, Fleming AM, White HS, Burrows CJ. Nanopore detection of 8-oxoguanine in the human telomere repeat sequence. ACS Nano. 2015;9(4):4296-307.
45. Oikawa S, Kawanishi S. Site-specific DNA damage at GGG sequence by oxidative stress may accelerate telomere shortening. FEBS Lett. 1999;453(3):365-8.
46. Wang Z, Rhee DB, Lu J, et al. Characterization of oxidative guanine damage and repair in mammalian telomeres. PLoS Genet. 2010;6(5):e1000951.
47. 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;33:1230–9.
48. 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.
49. McKinnon PJ. DNA repair deficiency and neurological disease. Nature reviews. Neuroscience. 2009;10:100–12.
50. Gillet LC, Scharer OD. Molecular mechanisms of mammalian global genome nucleotide excision repair. Chemical reviews. 2006;106:253–76.
51. Sugasawa K. Regulation of damage recognition in mammalian global genomic nucleotide excision repair. Mutation research. 2010;685:29–37.
52. 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;15:465–81.
53. de Laat WL, Jaspers NG, Hoeijmakers JH. Molecular mechanism of nucleotide excision repair. Genes Dev. 1999;13:768–85.
54. 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;92:258–62.
55. Rochette PJ, Brash DE. Human telomeres are hypersensitive to UV-induced DNA Damage and refractory to repair. PLoS Genet. 2010;6(4):e1000926.
56. Parikh D, Fouquerel E, Murphy CT, Wang H, Opresko PL. Telomeres are partly shielded from ultraviolet-induced damage and proficient for nucleotide excision repair of photoproducts. Nat Commun. 2015;6:8214.
57. Cordonnier AM, Fuchs RP. Replication of damaged DNA: molecular defect in xeroderma pigmentosum variant cells. Mutation research. 1999;435:111–9.

58. Pope-Varsalona H, Liu FJ, Guzik L, Opresko PL. Polymerase eta suppresses telomere defects induced by DNA damaging agents. Nucleic acids research. 2014;42:13096–109.
59. 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.
60. Harfe BD, Jinks-Robertson S. DNA mismatch repair and genetic instability. Annu Rev Genet. 2000;34:359–99.
61. Jiricny J. The multifaceted mismatch-repair system. Nat Rev Mol Cell Bio. 2006;7:335– 46.
62. 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;262:633–42.
63. Peltomaki P. Lynch syndrome genes. Fam Cancer. 2005;4:227–32.
64. Peltomaki P. Deficient DNA mismatch repair: a common etiologic factor for colon cancer. Hum Mol Genet. 2001;10:735–40.
65. 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.
66. 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;102:1300–5.
67. Mendez-Bermudez A, Royle NJ. Deficiency in DNA mismatch repair increases the rate of telomere shortening in normal human cells. Human mutation. 2011;32:939–46.
68. Campbell MR, Wang Y, Andrew SE, Liu Y. Msh2 deficiency leads to chromosomal abnormalities, centrosome amplification, and telomere capping defect. Oncogene. 2006;25:2531– 6.
69. Li GM. New insights and challenges in mismatch repair: getting over the chromatin hurdle. DNA repair. 2014;19:48–54.
70. Imai K, Yamamoto H. Carcinogenesis and microsatellite instability: the interrelationship between genetics and epigenetics. Carcinogenesis. 2008;29:673–80.
71. Ellegren H. Microsatellites: simple sequences with complex evolution. Nature reviews. Genetics. 2004;5:435–45.
72. Song N, Li Z, Qin N, et al. Shortened Leukocyte Telomere Length Associates with an Increased Prevalence of Chronic Health Conditions among Survivors of Childhood Cancer: A Report from the St. Jude Lifetime Cohort. Clin Cancer Res. 2020;26(10):2362-71.
73. Walsh KM. Telomere Attrition in Childhood Cancer Survivors. Clin Cancer Res. 2020;26(10):2281-3.
74. Walsh KM, Whitehead TP, de Smith AJ, et al. Common genetic variants associated with telomere length confer risk for neuroblastoma and other childhood cancers. Carcinogenesis. 2016;37(6):576-82.
75. El-Serag HB, Rudolph KL. Hepatocellular carcinoma: epidemiology and molecular carcinogenesis. Gastroenterology. 2007;132(7):2557–76.
76. Park JW, Chen M, Colombo M. et al. Global patterns of hepatocellular carcinoma management from diagnosis to death: the BRIDGE Study. Liver Int. 2015;35(9):2155–66.
77. Ding XX, Zhu QG, Zhang SM. et al. Precision medicine for hepatocellular carcinoma: driver mutations and targeted therapy. Oncotarget. 2017;8(33):55715–30.
78. Choi SH, Cho KJ, Yun SH, et al. HKR3 regulates cell cycle through the inhibition of hTERT in hepatocellular carcinoma cell lines. J Cancer. 2020;11(9):2442-52.
79. Shen Y, Xi F, Li H. et al. Telomerase reverse transcriptase suppression inhibits cell proliferation and promotes cell apoptosis in hepatocellular cancer. IUBMB Life. 2018;70(7):642– 8.