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ORIGINAL ARTICLE

Long telomeres at baseline and male sex are main determinants of telomere loss following chemotherapy exposure in lymphoma patients

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Abstract

Although chemotherapy (CHT) exposure is an established cause of telomere attrition, determinants of telomere length (TL) dynamics after chemotherapy are poorly defined. In this study, we analyzed granulocyte telomere dynamics in 34 adult lymphoma patients undergoing first-line CHT. TL was measured by southern blot at each CHT cycle and after 1 year from CHT completion. Median age was 59 yrs (range 22–77). Median number of CHT cycles was 6 (range 3–6). The majority of patients (79%, $n = 27$) experienced TL shortening following CHT exposure. Mean telomere loss was 673 base pairs (bp) by cycle 6. Telomere shortening was an early event as 87% of the total telomere loss (mean 586 bp) occurred by the end of cycle 3, with no significant recovery after 1 year. A significant correlation was observed between baseline TL and total or fractional telomere loss ($p < 0.001$), with telomere shortening by cycle 3 observed predominantly in male patients with long telomeres at pre-treatment evaluation. Stratifying the analysis by gender and age only young women (<51 years of age) did not show significant telomere shortening following chemotherapy exposure. These findings indicate that gender and baseline TL are major determinants of TL dynamics following chemotherapy exposure in lymphoma patients.

KEYWORDS
aging, chemotherapy, lymphoma, telomere, telomere loss

1 | INTRODUCTION

Telomeres are non-coding long tandem repeated sequences at the ends of chromosomes, which prevent accumulation of DNA damage and genomic instability.[1] Human telomerase (hTERT) is an enzyme able to restore telomere sequences thus preventing telomere shortening, and is commonly down-regulated in adults, leading to progressive telomere attrition with age.[2,3] In fact, leucocyte telomere length (TL) correlates with lifespan and has been extensively studied as a biomarker of ageing.[1] In adults, telomere erosion is more pronounced in men compared with women, and, accordingly, men have shorter life expectancy and higher cancer incidence.[1,4–7] Furthermore, several studies indicate that TL dynamics are altered in cancer, implying an important role of telomere attrition in cancer development and progression.[6–9] Oxidative and replicative stresses are among the most important mechanisms underlying telomere attrition in humans.[1] Several experimental systems of accelerated aging have been employed to investigate leucocyte telomere dynamics, including exposure to cytotoxic chemotherapy, autologous and allogeneic stem cell transplantation (ASCT and allo-SCT).[10–16] These models enabled the identification of intrinsic (e.g., genetic) and extrinsic factors influencing TL in vivo. Extrinsic modifiable factors are of special importance representing possible targets for therapeutic intervention.

Chemotherapy exposure, through direct and reactive oxygen species (ROS)-mediated DNA damage, is a well-established cause of telomere attrition.[1,14–17] Previous studies uncovered the relevance of telomere shortening in the setting of high dose chemotherapy and ASCT demonstrating that a reduced TL of the grafted cells could be an important predisposing factor for the development of secondary myeloid neoplasms.[18,19] However, few studies investigated determinants of TL dynamics following chemotherapy exposure in lymphoma patients treated in the first-line setting, with conflicting results.[20,21]

2 | METHODS AND MATERIALS

In the present study, we studied telomere dynamics in a series of 34 lymphoma patients undergoing first-line chemotherapy at the Hematology Department of Mauriziano Hospital (Turin, Italy), from 2013 to 2014. TL was measured by southern blotting on granulocyte fractions before and after each chemotherapy cycle, and at least one year after the end of the induction therapy. All patients gave a written informed consent. This retrospective, non-interventional study, was approved by the institutional review board and carried out according to the Helsinki declaration. Main patients characteristics are detailed in Table 1. All patients underwent bone marrow biopsy at initial staging, and only patients with no evidence of bone marrow lymphoma infiltration were included in this analysis. TL was assessed on granulocyte fractions, obtained through a modified density gradient separation method (Ficoll-Paque, GE Healthcare).

TL was assessed by Southern blot analysis as previously described.[10,13] Briefly, 2 μg of DNA were digested by mixing with Hinf I (20 U) and Rsa I (20 U) for 2 h at 37°C, according to manufacturer's recommendations (Roche Diagnostic). Digested DNA fragments were then separated by 0.8% agarose gel electrophoresis in 1 TAE running buffer1X (pH 8.0).After electrophoresis, gels were transferred to a positively charged nylon membrane (Roche Diagnostic Mannheim), and then exposed for 10′ to UV light to fix DNA fragments. The TeloTAGGG Telomere Length Assay Kit (Roche Diagnostic, Mannheim, Germany, catalogue N° 12209136001) was employed for the hybridization phase, in accordance with the manufacturer's instructions. Membranes were prehybridyzed for 2 hours

TABLE 1 Main patients characteristics

Parameter	No. =
No. of patients	
all	34
female	16
male	18
Age (yrs), median (range)	59 (22 – 77)
female	60 (23–73)
male	57 (22–77)
	p-value for age 0.55 (M vs. F)
Chemotherapy schedule	
ABVD	5
BEACOPP	1
BENDAMUSTINE	2
Ox-DHA	3
MINE	1
R-CHOP	19
R-CVP	3
No. of cycles delivered, median (range)	6 (3–6)
3 cycles	4
4–6 cycles	30
Lymphoma subtype	
DLBCL	15
FL	10
MZL	2
MCL	1
HL	6

Abbreviations: ABVD, Doxorubicin, Bleomycin, Vinblastine, Dacarbazine; BEACOPP, Bleomycin, Etoposide, Doxorubicin, Cyclophosphamide, Vincristine, Procarbazine, Prednisone; DLBCL, Diffuse Large B-cell Lymphoma; F, female; FL, Follicular Lymphoma; HL, Hodgkin Lymphoma; M, male; MCL, Mantle Cell Lymphoma; MINE, Mesna, Ifosfamide, Mitoxantrone, Etoposide; MZL, Marginal Zone Lymphoma; No., number; OXDHA, Oxaliplatin, High-Dose ARA-C, Dexametasone; R-CHOP, Rituximab-Cyclophosphamide, Doxorubicin, Vincristine, Prednisone; R-CVP, Rituximab, Cyclophosphamide, Vincristine, Prednisone.

in the prehybridization solution at 62C° and then hybrydized for 3 hours in the same conditions adding 2 μL of the digoxigenin (DIG)-labeled probe specific for telomeric repeats. Following hybridization, filters were washed twice at room temperature for 15 min in 2X washing solution and then twice at 39C° in 0.5X washing solution for 20 min. Filters were then incubated with a DIG-specific antibody covalently coupled to alkaline phosphate (AP Finally, results were visualized using AP metabolizing CDP-Star a highly sensitive chemiluminescent substrate. The light signal produced on the site of the hybridized probe was recorded on X-ray films (Lumi-Film Chemiluminescent Detection Film, Roche Diagnostic, Mannheim, Germany). Chemiluminescence is detected by x-ray film (RocheDiagnostics) and scanned for analysis. Analysis is performed using Quantity One 4.6.3 (Bio-Rad Laboratories). Telomeres are visualized as smears and the software calculates the intensity curve corresponding to each sample and the point of maximum signal intensity defined the highest concentration of telomeric repeats ('peak TRF length'). TRF length was determined by comparing the signals relative to standard molecular weights. Two different molecular weight standards were used in order to have a more precise estimate of the telomere length data obtained.

TL measurements were performed pretreatment at cycle 1 day 1 (C1D1), at C2D1, C3D1, C4D1, C5D1, C6D1, at day 30 after C6, and after at least 1 year from chemotherapy completion (+12–18 months).

2.1 | Statistical analysis

Patient characteristics were summarized by percentages, median values and ranges. Differences between groups were calculated with the Student *T* test and Mann-Whitney test. Box-plot graphs were used to show distributions of telomere length across different groups. The Pearson's test was used to establish correlations between different variables. Differences were considered significant for $p < 0.05$. Statistical analysis was performed with the statistical software package Prism 6.0 (GraphPad Software).

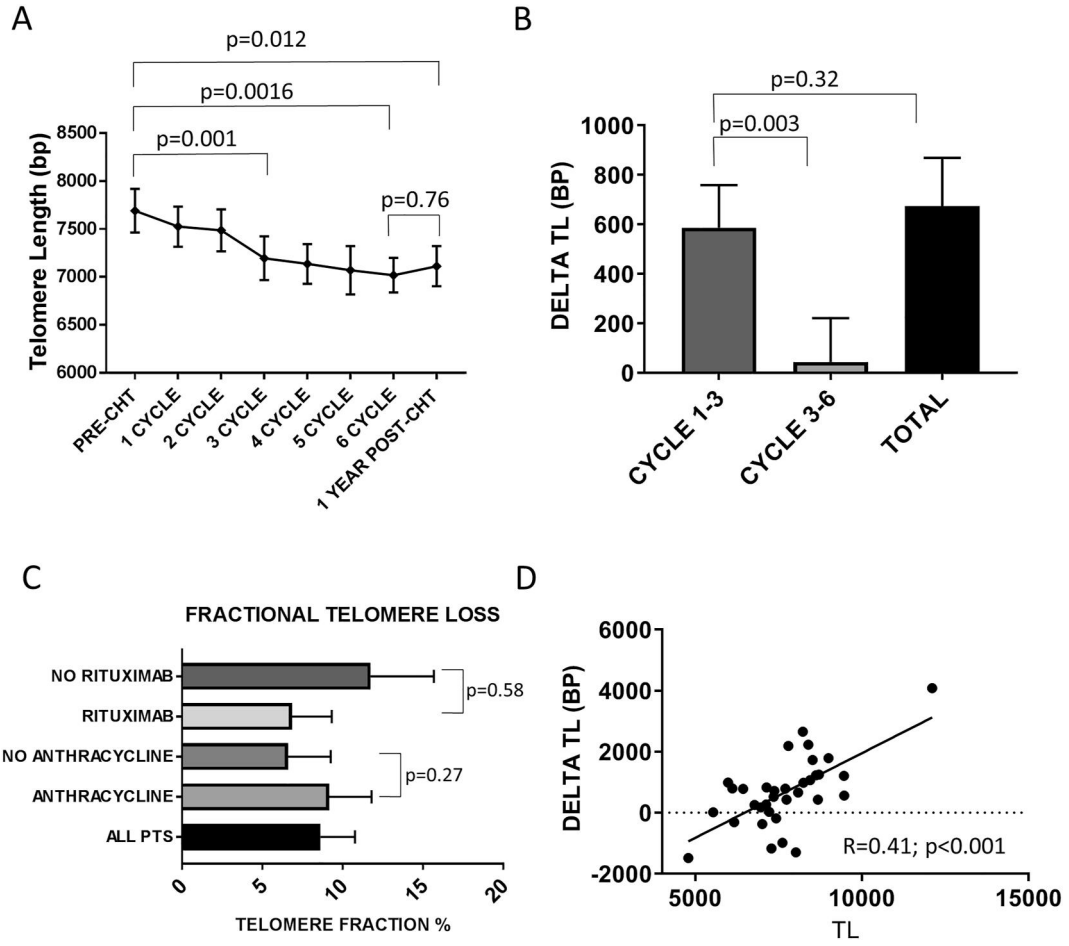

FIGURE 1 Characterization of granulocyte telomere length (TL) modifications in 34 lymphoma patients undergoing first-line chemotherapy treatment. (A). Changes in TL after 1,2,3,4,5,6 cycles of chemotherapy and after 1 year since therapy completion. Error bars represent standard error of the mean (S.E.M). *p* value was calculated with the paired *T* test. (B). Mean loss of TL in base pairs (DELTA TL, bp), after cycles 1-2-3, after cycles 4-5-6, and after all chemotherapy cycles (total). Error bars represent standard error of the mean (S.E.M). *p* value was calculated with the Mann-Whitney test. (C). Fractional telomere loss (telomere loss in bp/baseline TL) in the whole patient's population and in patients treated with or without anthracyclines or rituximab. Error bars represent standard error of the mean (S.E.M). *p* value was calculated with the Mann-Whitney test. (D). Linear correlation between degree of telomere loss (DELTA TL) and baseline TL, according to the Pearson's test.

3 | RESULTS

All patients considered in this study were undergoing their front-line treatment (Table 1). Median age was 59 yrs (range 22–77), with no significant differences between males and females ($p = 0.55$, Table 1). Overall, 26 patients (76%) were treated with antracycline-containing regimens. Rituximab immunotherapy was used in combination with chemotherapy in 22 patients (65%). Twenty-seven patients (79%) completed 6 cycles of therapy (median 6, range 3–6).

As expected, there was an inverse correlation between pre-treatment TL and age (p value = 0.01 $R = 0.18$, data not shown). Considering the whole patient's population, mean telomere loss by cycle 6 was 673 base pairs (bp) (8.7% of the pretreatment mean TL) (Figure 1A). Granulocyte TL decreased following chemotherapy exposure in 27 of 34 patients (79%), and in 25 of 27 patients total telomere loss exceeded 100 base pairs (bp). Among the latter group of patients, median telomere loss was 827 bp (range 177–4081). No significant differences in baseline characteristics were observed between patients experiencing telomere shortening ($n = 27$) and the 7 patients with increased telomere length after treatment (Table S1). Median increase in TL in the latter group assessed after chemotherapy completion (vs. pretreatment TL) was 986 bp (range 186–1482).

Telomere shortening was observed as early as after 1 cycle of chemotherapy, being significantly decreased after 3 and 6 cycles. Maximal drop in TL was observed at the end of induction therapy, with no significant recovery after 1 year (Figure 1A). Telomere shortening was an early event in the chemotherapy course, since 87% of the total telomere loss (mean 586 bp) occurred by the end of cycle 3, and only 13% from cycle 3 to cycle 6, as detailed in Figure 1B. Interestingly, maximal telomere erosion was determined by cycle 3 (Figure S1A and S1B). No significant differences in fractional telomere loss were observed in patients treated with chemotherapy regimens with or without anthracyclines or rituximab (Figure 1C). Representative southern blot analyses are shown in Figures S2 and S3.

Interestingly, we observed a significant correlation between baseline (pre-treatment) TL and total telomere loss, with a higher degree of telomere shortening following chemotherapy observed in those patients harboring the longest telomeres at pre-treatment evaluation (Figure 1D). In line with this observation, dividing patients into 2 subgroups based on the median baseline TL (7657 bp, range 4792–12104), significant telomere shortening was observed only among patients with longer telomeres at pre-treatment evaluation (Figure 2A). Importantly we did not observe significant differences with respect to baseline characteristics between the short and long telomere subgroups, with the exception that a higher fraction of long TL patients received anthracycline-based chemotherapy, compared to the short TL subgroup (Table 2). In light of this, we performed a separate analysis considering only anthracycline-treated patients, confirming increased telomere shortening in long TL patients, as compared to short TL patients (Figure S4). Overall, mean fractional telomere loss by cycle 6 was 14.4% of pretreatment TL in patients with long telomeres at baseline versus 0.3% in patients with short telomeres ($p = 0.0008$) (Figure 2B). Accordingly, while telomere shortening was negligible in patients with short telomeres, patients with long telomeres experienced a significant telomere loss by cycle 3 (Figure 2C). Notably, this trend was significantly more pronounced in male compared to female patients: in fact, in the long telomere subgroup only male patients experienced significant telomere shortening by cycle 3 (mean 1145 bp) (12.8% of the pretreatment mean TL), as compared to females, where total telomere loss (mean 205 bp) reached only 2.4% of the pretreatment mean TL (Figure 2C).

Following these observations, in order to ascertain whether the female milieu had an influence on telomere dynamics following chemotherapy, we stratified TL analyses by gender and age. Interestingly, among all patient's subgroups, only young women ≤51 years of age, which is the mean menopausal age in Europe,[22] did not show a significant decrease in TL following chemotherapy exposure (Figure 2D).

After a median follow-up of 6 years (range 1–8), no tAML/MDS cases were observed in this cohort. However, two patients were diagnosed with solid tumors, with pancreatic and bladder cancer occurring respectively after 4 and 3 years from chemotherapy completion. Maximal telomere loss was 1044 bp and 791 bp in the first and second case respectively. Overall, at the last follow-up, 3 patients had died: 2 patients in the short TL subgroup (DLBCL progression and pancreatic cancer respectively) and 1 patient in the long TL subgroup (complications of allo-SCT).

4 | DISCUSSION

In this study, we analyzed telomere dynamics in a cohort of lymphoma patients undergoing first line chemotherapy, providing some important observations: 1-the majority of patients experienced telomere attrition following chemotherapy; 2- telomere shortening was an early event, since the vast majority of telomere loss was accumulated during the first 3 chemotherapy courses; 3- persistent telomere loss was detected at 1 year since chemotherapy completion; 4- pre-treatment long telomeres were more prone to chemotherapy-induced telomere attrition; 5- highest telomere damage was observed in male patients with long telomeres at baseline; 6-significant telomere loss was observed in all patients subgroups with the exception of pre-menopausal women.

The observation that more than 80% of telomere loss is accumulated relatively early during chemotherapy is in line with similar findings in the setting of early-life telomere dynamics of humans and vertebrates, where accelerated telomere loss is commonly observed during the first years of age, when telomeres are longer, as compared to older ages.[23] On the other hand, we observed that the extent of telomere loss was strictly correlated with baseline telomere length, with pronounced telomere loss observed only in patients harboring long telomeres as compared to those with short telomeres. Thus, early and preferential attrition of long telomeres seems to be a common feature in different experimental models of aging.

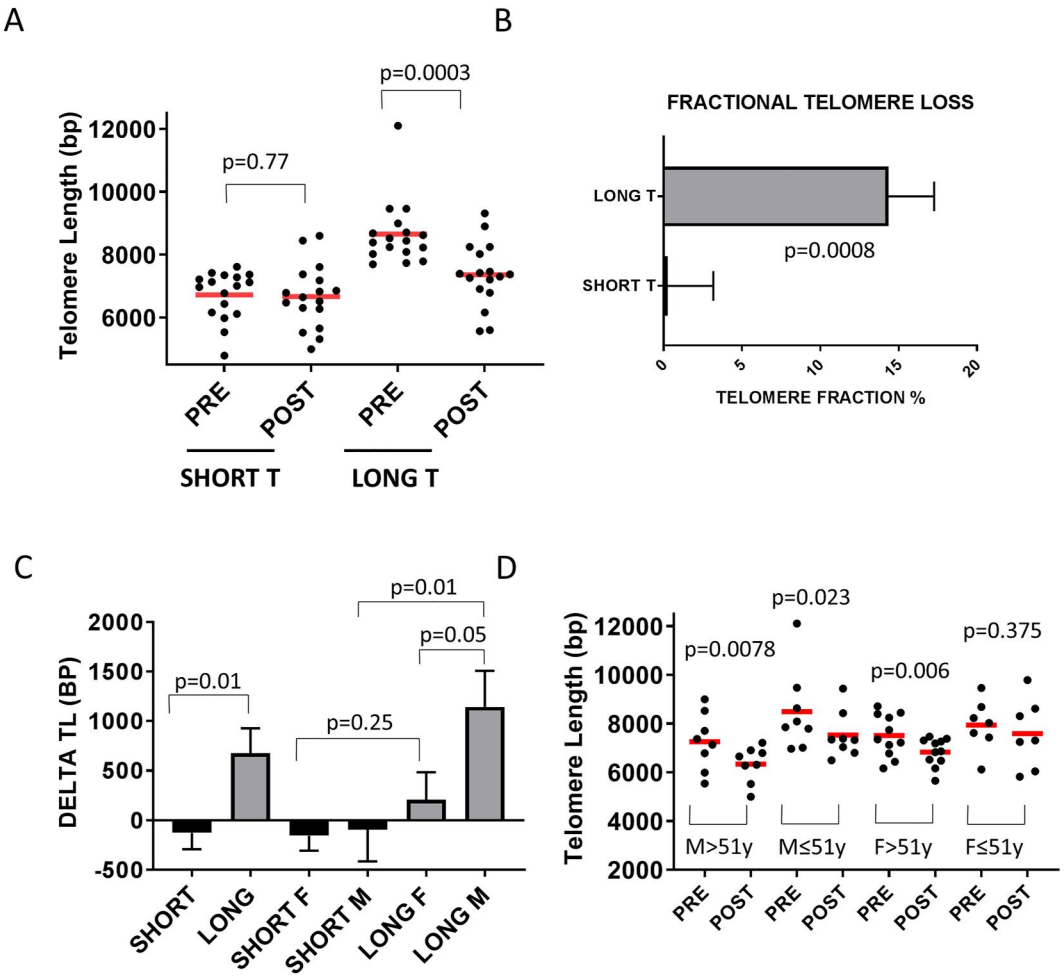

FIGURE 2 Impact of baseline telomere length (TL) and sex on telomere dynamics in 34 lymphoma patients undergoing first-line chemotherapy treatment. (A). Dot plot graph representing TL before and after chemotherapy in patient subgroups categorized according to baseline short (SHORT T) and long telomeres (LONG T). Patients were divided in the 2 subgroups based on the median TL. Differences between groups were calculated with the paired *T* test. (B). Fractional telomere loss (telomere loss in bp/baseline TL) in patients with long and short telomeres at baseline. Error bars represent standard error of the mean (S.E.M). *p* value was calculated with the Mann-Whitney test. (C). Mean loss of TL in base pairs (DELTA TL, bp) from cycle 1 to 3 according to baseline TL (SHORT vs. LONG) and sex Male (M) versus Female (F). Error bars represent standard error of the mean (S.E.M). *p* value was calculated with the Mann-Whitney test. (D). Pre and post chemotherapy TL in patient subgroups categorized according to age (>51 y.o vs. </ = 51 y.o) and Male (M) or Female (F) gender. Differences between groups were calculated with the paired *T* test

Interestingly, a recent systematic review of 25 studies could not draw definite conclusions on the effects of chemotherapy and radiotherapy on leukocyte TL in unselected patients populations.[20] Our study clearly demonstrates that the extent of telomere attrition is not homogeneous across all patient subsets, and that specific patient populations could be at higher risk of telomere loss following chemotherapy, such as patients with long telomeres at baseline. Whether these observations indicate an increased susceptibility to oxidative damage in cells with long telomeres, or a selection of subclones with short telomeres following exposure to chemotherapy, is a very relevant question which should be addressed in future studies. Irrespective of the underlying mechanisms, the preferential loss of long telomeres was significantly more pronounced in male patients compared to females. Furthermore, leukocyte TL was relatively preserved in pre-menopausal women. These findings are in line with previous observation from our group[13] and others,[5,24-26] pointing to a protective role of the female milieu on telomere maintenance dynamics. Since rate of telomere loss has been associated with life-span,[27] and telomere attrition after chemotherapy has been associated with an increased risk of therapy-related acute myeloid leukemia or myelodysplasia (t-AML/MDS),[18,19,28] these data could have relevant clinical implications providing the rationale for the use of female hormones as anti-aging agents, and to prevent tAML/MDS in selected high-risk patients subgroups. Due to a relatively short follow-up and considering the small number of patients included, we did not observe any tAML/MDS case in this cohort; however, 2 of 34 patients (5.9%) were diagnosed with secondary solid tumors. Interestingly both patients experienced telomere loss

TABLE 2 Patients characteristics in the short and long telomere lenght subgroups

Parameter	Short TL subgroup	Long TL subgroup	*p*-Value
No. of patients			
all	17	17	-
female	9	9	
male	8	8	
Baseline TL (bp), median (range)	7007 (4792–7615)	8444 (7698–12104)	<0.0001
Age (yrs), median (range)	64 (27 – 73)	55 (22–77)	0.31
Chemotherapy schedule			
ABVD	2	3	-
BEACOPP	0	1	-
BENDAMUSTINE	1	1	-
Ox-DHA	3	0	-
MINE	0	1	-
R-CHOP	8	11	-
R-CVP	3	0	-
Anthracycline			0.03
Yes	10	16	
No	7	1	
Rituximab			
Yes	11	11	-
No	6	6	
No. of cycles delivered, median (range)	6 (3–6)	6 (3–6)	-
3 cycles	2	2	
4–6 cycles	15	15	
Lymphoma subtype			
NHL	15	13	0.65
DLBCL	5	10	0.16
FL	7	3	0.25
MZL	2	0	0.48
MCL	1	1	-
HL	2	4	0.65

Abbreviations: ABVD, Doxorubicin, Bleomycin, Vinblastine, Dacarbazine; BEACOPP, Bleomycin, Etoposide, Doxorubicin, Cyclophosphamide, Vincristine, Procarbazine, Prednisone; DLBCL, Diffuse Large B-cell Lymphoma; F, female; FL, Follicular Lymphoma; HL, Hodgkin Lymphoma; M, male; MCL, Mantle Cell Lymphoma; MINE, Mesna, Ifosfamide, Mitoxantrone, Etoposide; MZL, Marginal Zone Lymphoma; No., number; OXDHA, Oxaliplatin, High-Dose ARA-C, Dexametasone; R-CHOP, Rituximab-Cyclophosphamide, Doxorubicin, Vincristine, Prednisone; R-CVP, Rituximab, Cyclophosphamide, Vincristine, Prednisone.

after chemotherapy exposure. Notably, recent studies confirm a mechanistic role of telomere shortening in immune-mediated inflammatory dysfunction and cancer initiation in preclinical models.[29] Subgroup analyses addressing the long-term effects of telomere attrition on tAML/MDS incidence and cancer immune surveillance in general, should be the goal of future studies with longer follow-up, bigger sample size, and external validation cohorts. These studies should be focusing on the relative risk of secondary malignancies in long TL patients at baseline experiencing significant treatment-related telomere shortening versus short TL patients with negligible telomere loss following therapy.

5 | CONCLUSIONS

This characterization of telomere dynamic following in vivo exposure to chemotherapy may offer some useful insights for understanding the physiological process leading to cell ageing. Baseline TL and female milieu emerged as powerful determinants of telomere dynamics following chemotherapy exposure, thus indicating that telomere maintenance is a complex process influenced by a combination of intrinsic and extrinsic, (thus modifiable), factors.

These observations, together with our previous findings on the effects of female hormones on telomere maintenance in the transplant setting,[13] could pave the way for future telomere-protective therapies in specific high-risk patient's subsets following chemotherapy exposure.

AUTHOR CONTRIBUTIONS

Enrico Derenziniand Corrado Tarella conceived the study, analyzed the data and wrote the manuscript; Angela Gueli, Riccardo Bruna, Daniela Gottardi, Alessandro Cignetti collected clinical data and revised the manuscript; Alessandra Risso performed southern blot experiments; Enrico V. Avvedimento conceived the study and critically revised the manuscript.

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CONFLICT OF INTEREST

ED received research funding from TG-Therapeutics, ADC-Therapeutics, Takeda and served in the Advisory board for Gilead, Astra Zeneca, Takeda and Beigene. CT served in the advisory board of ADC- Therapeutics and AbbVie.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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PEER REVIEW

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REFERENCES

1. Blackburn EH, Epel ES, Lin J. Human telomere biology: a contributory and interactive factor in aging, disease risks, and protection. *Science*. 2015;350(6265):1193-1198. https://doi.org/10.1126/science.aab3389
2. Blackburn EH, Collins K. Telomerase: an RNP enzyme synthesizes DNA. *Cold Spring Harb Perspect Biol*. 2011;3(5):a003558. https://doi.org/10.1101/cshperspect.a003558
3. Collins K, Mitchell J. Telomerase in the human organism. *Oncogene*. 2002;21(4):564-579. https://doi.org/10.1038/sj.onc.1205083
4. Barrett EL, Richardson DS. Sex differences in telomeres and lifespan. *Aging Cell*. 2011;10(6):913-921. https://doi.org/10.1111/j.1474-9726.2011.00741.x
5. Mayer S, Brüderlein S, Perner S, et al. Sex-specific telomere length profiles and age-dependent erosion dynamics of individual chromosome arms in humans. *Cytogenet Genome Res*. 2006;112(3-4):194-201. https://doi.org/10.1159/000089870
6. Kim NW, Piatyszek MA, Prowse KR, et al. Specific association of human telomerase activity with immortal cells and cancer. *Science*. 1994;266(5193):2011-2015. https://doi.org/10.1126/science.7605428
7. Telomeres Mendelian Randomization Collaboration, Haycock PC, Burgess S, et al. Association between telomere length and risk of cancer and non-neoplastic diseases: a mendelian randomization study. *JAMA Oncol*. 2017;3(5):636-651. https://doi.org/10.1001/jamaoncol.2016.5945
8. Barthel FP, Wei W, Tang M, et al. Systematic analysis of telomere length and somatic alterations in 31 cancer types. *Nat Genet*. 2017;49(3):349-357. https://doi.org/10.1038/ng.3781
9. Kroupa M, Rachakonda SK, Liska V, et al. Relationship of telomere length in colorectal cancer patients with cancer phenotype and patient prognosis. *Br J Cancer*. 2019;121(4):344-350. https://doi.org/10.1038/s41416-019-0525-3
10. Ricca I, Compagno M, Ladetto M, et al. Marked telomere shortening in mobilized peripheral blood progenitor cells (PBPC) following two tightly-spaced high-dose chemotherapy courses with G-CSF. *Leukemia*. 2005;19(4):644-651. https://doi.org/10.1038/sj.leu.2403652
11. Ruella M, Rocci A, Ricca I, et al. Comparative assessment of telomere length before and after hematopoietic SCT: role of grafted cells in determining post-transplant telomere status. *Bone Marrow Transpl*. 2010;45(3):505-512. https://doi.org/10.1038/bmt.2009.297
12. Wynn RF, Cross MA, Hatton C, et al. Accelerated telomere shortening in young recipients of allogeneic bone-marrow transplants. *Lancet*. 1998;351(9097):178-181. https://doi.org/10.1016/S0140-6736(97)08256-1
13. Derenzini E, Risso A, Ruella M, et al. Influence of donor and recipient gender on telomere maintenance after umbilical cord blood cell transplantation: a study by the gruppo italiano trapianto di midollo osseo. *Biol Blood Marrow Transpl*. 2019;25(7):1387-1394. https://doi.org/10.1016/j.bbmt.2019.03.026
14. Lu Y, Leong W, Guerin O, Gilson E, Ye J. Telomeric impact of conventional chemotherapy. *Front Med*. 2013;7(4):411-417. https://doi.org/10.1007/s11684-013-0293-z
15. Buttiglieri S, Ruella M, Risso A, et al. The aging effect of chemotherapy on cultured human mesenchymal stem cells. *Exp Hematol*. 2011;39(12):1171-1181. https://doi.org/10.1016/j.exphem.2011.08.009
16. Alhareeri AA, Archer KJ, Fu H, et al. Telomere lengths in women treated for breast cancer show associations with chemotherapy, pain symptoms, and cognitive domain measures: a longitudinal study. *Breast Cancer Res*. 2020;22(1):137. https://doi.org/10.1186/s13058-020-01368-6
17. 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):2362-2371. https://doi.org/10.1158/1078-0432.CCR-19-2503
18. Chakraborty S, Sun CL, Francisco L, et al. Accelerated telomere shortening precedes development of therapy-related myelodysplasia or acute myelogenous leukemia after autologous transplantation for lymphoma. *J Clin Oncol*. 2009;27(5):791-798. https://doi.org/10.1200/JCO.2008.17.1033
19. Tarella C, Passera R, Magni M, et al. Risk factors for the development of secondary malignancy after high-dose chemotherapy and autograft, with or without rituximab: a 20-year retrospective follow-up study in patients with lymphoma. *J Clin Oncol*. 2011;29(7):814-824. https://doi.org/10.1200/JCO.2010.28.9777
20. Gallicchio L, Gadalla SM, Murphy JD, Simonds NI. The effect of cancer treatments on telomere length: a systematic review of the literature. *J Natl Cancer Inst*. 2018;110(10):1048-1058. https://doi.org/10.1093/jnci/djy189
21. Lee JJ, Nam CE, Cho SH, Park KS, Chung IJ, Kim HJ. Telomere length shortening in non-Hodgkin's lymphoma patients undergoing chemotherapy. *Ann Hematol*. 2003;82(8):492-495. https://doi.org/10.1007/s00277-003-0691-4
22. Meschia M, Pansini F, Modena AB, et al. Determinants of age at menopause in Italy: results from a large cross-sectional study. *ICARUS Study Group. Matur*. 2000;34(2):119-125. https://doi.org/10.1016/s0378-5122(99)00095-x
23. Frenck RW, Blackburn EH, Shannon KM. The rate of telomere sequence loss in human leukocytes varies with age. *Proc Natl Acad Sci USA*. 1998;95(10):5607-5610. https://doi.org/10.1073/pnas.95.10.5607
24. Brüderlein S, Müller K, Melzner J, Hogel J, Wiegand P, Moller P. Different rates of telomere attrition in peripheral lymphocytes in a pair of dizygotic twins with hematopoietic chimerism. *Aging Cell*. 2008;7(5):663-666. https://doi.org/10.1111/j.1474-9726.2008.00413.x
25. Viña J, Borrás C, Gambini J, Sastre J, Pallardó FV. Why females live longer than males? Importance of the upregulation of longevity-associated genes by oestrogenic compounds. *FEBS Lett*. 2005;579(12):2541-2545. https://doi.org/10.1016/j.febslet.2005.03.090
26. Calado RT, Yewdell WT, Wilkerson KL, et al. Sex hormones, acting on the TERT gene, increase telomerase activity in human primary hematopoietic cells. *Blood*. 2009;114(11):2236-2243. https://doi.org/10.1182/blood-2008-09-178871
27. Whittemore K, Vera E, Martínez-Nevado E, Sanpera C, Blasco MA. Telomere shortening rate predicts species life span. *Proc Natl Acad Sci USA*. 2019;116(30):15122-15127. https://doi.org/10.1073/pnas.1902452116
28. Morton LM, Dores GM, Schonfeld SJ, et al. Association of chemotherapy for solid tumors with development of therapy-related myelodysplastic syndrome or acute myeloid leukemia in the modern era. *JAMA Oncol*. 2019;5(3):318-325. https://doi.org/10.1001/jamaoncol.2018.5625
29. Lex K, Maia Gil M, Lopes-Bastos B, et al. Telomere shortening produces an inflammatory environment that increases tumor incidence

in zebrafish. *Proc Natl Acad Sci U S A*. 2020;117(26):15066-15074. https://doi.org/10.1073/pnas.1920049117

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

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