
HUMAN
GENETICS

Individual Differences in Relative Telomere Length in Mentally Healthy Subjects: The Effect of *TERT* Gene Polymorphism and Urban Residency

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Received January 11, 2022; revised March 17, 2022; accepted April 4, 2022

Abstract—The changes in the telomere length caused by the terminal underreplication in the existing literature are related to depressive disorders. However, the use of the telomere length as a biomarker of depressive states is ambiguous, which is due to the effect of various environmental factors on both the psychoemotional state and cellular aging of an organism. In order to identify the possible use of the relative telomere length (RTL) measured in peripheral blood leukocytes as a biomarker of enhanced liability to depression prior to the clinical symptoms, as well as to determine the link between telomere length, sociodemographic factors, allelic variants of the genes involved in the regulation of telomere elongation, and depression level, the association analysis of reverse transcriptase (*TERT* rs7726159), telomerase RNA component (*TERC* rs1317082), and the CST complex encoding protein (*OBFC1* rs2487999) gene polymorphisms was performed with RTL and depression level in mentally healthy individuals ($N = 1065$) aged 18–25 years. Together with genetic variants, the examined regression models included various sociodemographic parameters as predictors. As a result of statistical analysis, we failed to observe the association between RTL and individual differences in depression level in the studied sample. Nevertheless, multiple regression analysis allowed us to construct a statistically significant model of individual variance in RTL ($P = 4.3e-4$; $r^2 = 0.018$), which included rs7726159 in the *TERT* gene ($P = 0.020$; $\beta = 0.078$) and such environmental predictors as age ($P = 0.001$; $\beta = -0.027$) and place of residence in childhood (urban/rural area) ($P = 0.048$; $\beta = 0.063$). The data obtained confirm the involvement of *TERT* gene variants and age in telomere length in mentally healthy individuals aged 18–25 years and indicate a negative effect of urban residency on telomere length shortening, which reflects the cellular aging of an organism.

Keywords: telomeres, telomerase, telomerase RNA component, stress, depression, urban residency, SNP

DOI: 10.1134/S1022795422090101

INTRODUCTION

Telomeres (telomeric repeats) are heterochromatin structures located at the ends of chromosomes, necessary to maintain their integrity and stability [1]. It is known that telomeres consist of 5'-TTAGGG-3' tandem repeats with a length of 10–15 kb in humans, which inevitably decrease at a rate of 50 to 200 bp during each replication cycle owing to the so-called “terminal underreplication problem” and the action of specific nucleases [2]. Nevertheless, this problem is solved in cells by a compensating effect of telomerase—ribonucleic reverse transcriptase. Telomerase

consists of two main components, namely, reverse transcriptase (encoded by the *TERT* gene, 5p15.33) and telomerase RNA component (encoded by the *TERC* gene, 3q26.2) containing a matrix site for the synthesis of telomeric repeats [3]. In a healthy human body, telomerase is highly active in germ and stem cells and exhibits moderate activity in leukocytes [3]. On the other hand, the length of telomeric repeats is regulated by shelterin and CST complexes [4]. The CST complex in higher eukaryotes consists of Stn1 (encoded by the *OBFC1* gene), Ten1, and CTC1 pro-

teins and participates in the inhibition of telomerase-dependent elongation of telomeric repeats [4].

Twin studies indicate the heritability of telomere length ~36%, while environmental factors determine ~49% of changes in their length during ontogenesis [5]. To date, genome-wide association studies (GWAS) have identified single-nucleotide polymorphisms (SNPs) in the *TERC*, *TERT*, and *OBFC1* genes, whose association with telomere length has been replicated [6, 7]. A functional significance has been shown for *TERT* rs7726159, *TERC* rs1317082, and *OBFC1* rs2487999 associated with telomere length [7].

Changes in the length of telomeric repeats, measured in absolute or relative units, in the existing literature are related to the development of oncological diseases [7], cardiovascular pathology [8], and several mental disorders, including post-traumatic stress disorder [3], depression [9], and Alzheimer's disease [10]. It is assumed that one of the possible molecular mechanisms underlying the development of mental disorders is attributed to the effect of a shorter telomere length on the functioning of immune cells in the nervous system or in the blood circulatory system, which increases inflammatory processes in the central nervous system [10].

According to the published data, a measurement of the relative telomere length in easily accessible samples such as peripheral blood can provide reliable data for correlation of a relative telomere length in other tissues, including neuronal [11]. The possibility of using the relative telomere length (RTL) in peripheral blood leukocytes as a biomarker of depressive disorders is ambiguous: some studies report shortening of telomeres in patients with depression compared to the control group [1, 9], while other studies failed to observe such a link [12].

Together with a traditional approach, the use of bioinformatic pipelines to estimate telomere length under whole-genome sequencing (TelSeq) revealed that shortening of telomeres in depressive disorders was associated with adverse environmental conditions [1]. It is known that adverse stress conditions at an early age cause a pleiotropic behavioral, physiological, and epigenetic effect on various systems of the body [13] owing to the changes in the stress response of the hypothalamic-pituitary-adrenal (HPA) system, which, in turn, can result in impaired mental health disorders [14, 15]. Environmental factors affecting telomere length include age-dependent decrease in RTL [8], specificity of child–parent relations [16], adverse traumatic events in childhood [15], stressful life effects [17], environmental influence [18], and the level of family income [19]. It should be noted that men, compared to women, and individuals of European origin, compared to individuals of other ethnicity, are characterized by shorter telomere length [19]. The accumulation of age-dependent reactive oxygen species and free radicals in the cells, which is associated with the

effects of stress factors, inflammatory reactions, and unhealthy lifestyle (smoking, excessive alcohol intake, low physical activity), results in shortening of telomeres [2, 18]. Considering a possible effect of the aforementioned environmental factors on the emotional state [20, 21] and cellular aging of the body, the study of a large array of sociodemographic characteristics in the context of their effect on telomere length is of relevance.

Therefore, in the present study we suggested that the relative length of telomeric repeats could be used as a biomarker, which reflects the effect of adverse environmental factors at previous stages of human ontogenesis and/or individual stress sensitivity, which, in turn, could determine a manifestation of depression. An alternative hypothesis suggested that telomere length depended on individual stress sensitivity and emotional state together with certain environmental factors and genetic variants associated with telomerase action and the CST complex.

MATERIALS AND METHODS

The study included 1065 mentally healthy individuals (79% women)—students at the universities of the Republic of Bashkortostan and the Udmurt Republic (mean age 20.99 ± 3.32 years) of different ethnic origin, including 357 Russians, 340 Tatars, 234 Udmurts, and 134 individuals of mixed ethnicity. All enrolled individuals had no individual and familial history of mental disorders. The participants were subjected to psychological testing of depression level using Beck Depression Inventory (BDI), which was validated in the Russian population. In addition, all the subjects filled out a questionnaire to obtain the data on ethnic background up to three generations and specificity of rearing and parent–childhood relations, including the episodes of childhood maltreatment, rearing in a complete/incomplete family, family income, maternal age at delivery, place of residence (urban/rural), number of children in a family, and individual weight at birth (weight under 2500 g was considered as low). The place of residence (urban/rural residency) was determined on the basis of the population of the locality: demographic units with a population less than 50 000 people were assigned to rural areas [21]. On the basis of the survey, we formed the database consisting of information on birth order and sibship size (number of children in a family), rearing style (according to the Parental Bonding Inventory), and rearing in a bilingual family for individuals of Tatar and Udmurt origin. An informed consent for the voluntary involvement in the present study was obtained from all the participants. This study was approved by the Bioethics Committee at the Institute of Biochemistry and Genetics, Ufa Federal Research Centre, Russian Academy of Sciences.

A collection of biological material (peripheral blood) was carried out in 2017–2019, followed by

DNA isolation via phenol-chloroform extraction. Genotyping of *TERT* rs7726159, *TERC* rs1317082, and *OBFC1* rs2487999 polymorphisms was performed under a technology of concurrent allele-specific PCR (KASP™, LGC Genomics, UK) on a CFX96 DNA Analyzer (BioRad, United States), which provides the endpoint collection of fluorescence data. The results of distribution of allele and genotype frequencies of the examined polymorphic loci corresponded to the Hardy–Weinberg equilibrium: $P = 0.84$ for rs7726159 (*TERT*), $P = 0.63$ for rs1317082 (*TERC*), and $P = 0.56$ for rs2487999 (*OBFC1*). To estimate the relative telomere length in DNA of examined samples, the concentration of the samples was aligned to 20 ng/μL for the subsequent analysis according to the study [22]. Detection of synthesized product was conducted using intercalating fluorescent dye IQ SYBR Green Supermix (BioRad). Real-time PCR (CFX96, BioRad) was based on the primers for the amplification of telomere repeats (T) and a single-copy β-globin gene (*HGB*), which served as a conservative gene (S) [23]. During real-time PCR, a threshold cycle (Ct) in the case of amplification of a conservative gene (S) and DNA fragment consisting of telomere repeats was registered. Since each sample (both experimental and standard one) was run in triplicate, an average cycle threshold for each sample was calculated. If the differences in Ct values between the triplicates exceeded 30%, the sample was excluded from the analysis. Pool DNA consisting of DNA from several individuals served as a control sample and was identical in each run. Reproducibility of the results was above 98%.

To determine the relative telomere length, we used the widely accepted technique [22] based on the formula $2^{-\Delta\Delta Ct}$. For this purpose, the difference in the cycle thresholds between the telomere and control PCR and the relative telomere length in a genome (T/S) were calculated according to the formula $T/S = 2^{-\Delta\Delta Ct}$, where $\Delta\Delta Ct = (Ct_T(\text{sample}) - Ct_T(\text{poolDNA})) - (Ct_S(\text{sample}) - Ct_S(\text{poolDNA}))$. According to published data, the relative telomere length in a genome (T/S) correlated with $2^{-\Delta\Delta Ct}$ and telomere length in the examined sample.

To estimate the link between the relative telomere length and phenotypic variance in depression level, we carried out a correlation analysis using Spearman's rank correlation coefficient and multiple linear regression analysis (RStudio v4.1.2). The level of depression (in points) or the relative telomere length served as a dependent variable, while RTL (or depression level, respectively) together with *TERT*, *TERC*, and *OBFC1* gene SNPs and environmental factors were included in the model as independent predictors. In order to obtain the model with the highest predictive significance, we used a backward elimination algorithm, which dropped all insignificant parameters until the achievement of the best parameters in the model including variance level (effect size, r^2), significance

level (p -value), and Akaike criterion. The level of statistical significance was accepted at 0.05.

RESULTS

Within the framework of the present study, we conducted a correlation and linear regression analysis between the level of depression and the relative telomere length in peripheral blood leukocytes in individuals aged 18–25 years, which failed to detect a correlation ($P = 0.465$; $r = -0.023$) and association ($P = 0.155$; $\beta = -0.003$) between these parameters. Since the published data point to the presence of differences in RTL depending on sex and ethnic origin, we performed an estimate of differences in RTL between the groups. No statistically significant differences were observed between men and women ($P = 0.137$) or between individuals of Russian and Tatar ethnicity ($P = 0.428$). At the same time, statistically significant differences in RTL were determined between Russians and Udmurts ($P = 0.025$) and Tatars and Udmurts ($P = 0.005$). Therefore, subsequent linear regression analysis directed to the assessment of the effect of SNPs in the *TERT* rs7726159, *TERC* rs1317082, and *OBFC1* rs2487999 gene on individual differences in telomere length was carried out with inclusion of ethnicity as a covariate. As a result of analysis, statistically significant differences in RTL were confirmed between carriers of *TERT* rs7726159*A/C genotype and major rs7726159*C/C genotype in the additive model ($P = 0.015$; $\beta = 0.076$; $r^2 = 0.004$) (Tables 1, 2). In particular, RTL shortening was characteristic of individuals not having the minor rs7726159*A allele (Table 1, Fig. 1a). The mean values of RTL with respect to genotypes of the studied SNPs in the total sample and among men, women, Russians, Tatars, and Udmurts are shown in Table 1. At the same time, no association between *TERC* rs1317082 ($P = 0.893$) and *OBFC1* rs2487999 ($P = 0.426$) and RTL in the examined sample was observed. As a result of the use of backward elimination algorithm, we revealed a statistically significant model of interindividual differences in RTL ($P = 5.9e-4$; $r^2 = 0.015$), which included age ($P = 0.001$; $\beta = -0.027$) and place of residence ($P = 0.066$; $\beta = 0.059$) (Fig. 1b). Simultaneous inclusion of genetic and environmental predictors allowed us to determine the most significant model ($P = 4.3e-4$; $r^2 = 0.018$), including *TERT* rs7726159 ($P = 0.019$; $\beta = 0.078$), age ($P = 0.001$; $\beta = -0.027$), and place of residence ($P = 0.048$; $\beta = 0.063$) (Table 2; Figs. 1c, 1d).

In order to verify a hypothesis of relation of depression level to variance in telomere length and examined genetic variants, we conducted a series of multiple regression analyses, which failed to detect the association between *TERT* rs7726159 ($P = 0.239$), *TERC* rs1317082 ($P = 0.768$), *OBFC1* rs2487999 ($P = 0.110$), and RTL ($P = 0.195$) (Table 2) and psychological traits in mentally healthy individuals without clinical symptoms of depression.

Table 1. Mean values (standard deviation) of relative telomere length in the examined groups with respect to *TERT* rs7726159, *TERC* rs1317082, and *OBFC1* rs2487999 genotypes

SNP	Group	Total sample (<i>N</i> = 1065)	Women (<i>N</i> = 841)	Men (<i>N</i> = 224)	Russians (<i>N</i> = 357)	Tatars (<i>N</i> = 340)	Udmurts (<i>N</i> = 234)
<i>TERT</i> rs7726159	<i>C/C</i>	0.94 ± 0.46 ^a	0.94 ± 0.45 ^b	0.97 ± 0.47	0.96 ± 0.44	0.89 ± 0.47 ^c	0.98 ± 0.40
	<i>A/C</i>	1.03 ± 0.50	1.05 ± 0.52	0.93 ± 0.40	1.02 ± 0.52	1.01 ± 0.47	1.12 ± 0.54
	<i>A/A</i>	1.00 ± 0.44	1.02 ± 0.42	0.90 ± 0.52	0.89 ± 0.40	1.03 ± 0.51	1.12 ± 0.41
<i>TERC</i> rs1317082	<i>A/A</i>	0.99 ± 0.46	1.00 ± 0.46	0.93 ± 0.45	0.99 ± 0.48	0.94 ± 0.42	1.08 ± 0.44
	<i>A/G</i>	0.99 ± 0.48	0.99 ± 0.48	0.98 ± 0.45	0.98 ± 0.48	0.98 ± 0.52	1.06 ± 0.41
	<i>G/G</i>	0.96 ± 0.54	0.99 ± 0.57	0.86 ± 0.41	0.99 ± 0.45	0.94 ± 0.47	1.01 ± 0.66
<i>OBFC1</i> rs2487999	<i>C/C</i>	1.00 ± 0.48	1.01 ± 0.49	0.93 ± 0.43	0.99 ± 0.48	0.97 ± 0.48	1.07 ± 0.47
	<i>C/T</i> + <i>T/T</i>	0.96 ± 0.45	0.94 ± 0.44	1.02 ± 0.53	0.97 ± 0.44	0.90 ± 0.45	0.99 ± 0.45

Statistically significant differences between the groups (Kruskal–Wallis test): ^a $\chi^2 = 7.03$, $P = 0.030$; ^b $\chi^2 = 9.22$, $P = 0.010$; ^c $\chi^2 = 7.48$, $P = 0.024$.

DISCUSSION

In the present study, we failed to confirm our initial hypothesis on the link between depression level in mentally healthy young adults and telomere length with inclusion of environmental factors and SNPs in the genes which regulate restoration of telomere length as predictors. Nevertheless, within the framework of the present study, the place of residence (urban/rural residency), age, and the number of minor alleles of *TERT* rs7726159 were demonstrated to predict individual differences in telomere length. The results of published GWAS are congruent with our findings, thus indicating the association of minor allele of *TERT* rs7726159 with enhanced telomere length [7]. Despite the published findings on the relation of shortening of telomeres to the presence of a minor allele of *TERC* rs1317082 and their increase with a minor allele of *OBFC1* rs2487999 [7], we failed to detect such associations. Despite the declared hypothesis on the association between telomere length and depression level and published data on the effect of *TERT* and *TERC* SNPs in manifestation of internalizing problems (depression, anxiety, and post-traumatic stress disorder) [3, 24], we did not confirm these associations between genetic variants and depression level in mentally healthy young adults.

According to the results of previous research, all three examined SNPs (*TERT* rs7726159, *TERC* rs1317082, *OBFC1* rs2487999) have been identified in GWAS of telomere length [6, 7]. Our findings indicate the effect of only *TERT* rs7726159 (the main effect and gene-by-environment interactions) in individual differences in telomere length in mentally healthy young adults. In particular, carriers of the heterozygous rs7726159**A/C* genotype were characterized by enhanced RTL compared to carriers of the major rs7726159**C/C* genotype. Previously, in a large sample ($N = 66363$) of individuals from the Danish popu-

lation, a statistically significant shortening of telomeres in carriers of the rs7726159**C* allele in the *TERT* gene was observed [10], which is congruent with our findings also reporting a decreased RTL in individuals bearing the rs7726159**C/C* genotype compared to carriers of the minor *A* allele.

Several published studies which simultaneously considered both the role of factors reflecting the environmental effect in childhood and telomere length on developing postpartum depression reported no association between depression level and variations in telomere length [15]. At the same time, adverse environmental impacts in childhood play a significant role in changes in telomere length and modifications in the profile of DNA methylation, indicating an increase in the allostatic load of many systems of the body [15]. Other researchers evaluating a possible role of depression in the changes in telomere length within a large-scale study of depression and anxiety (Netherlands Study of Depression and Anxiety) demonstrated the absence of a modifying effect of negative psychoemotional state on the number of telomere repeats, which is congruent to our findings, although they observed the influence of an adverse environment on the shortening of telomeres [18]. It should be noted that constantly increased stress level promotes a significant shortening of chromosomal telomeres in a longitudinal paradigm, and sex specificity in the perception of stress factors of various nature has been shown [17].

The influence of age on telomere length has been discussed for a long time [2, 18]. It is known that telomere length is diminished in normal cells with age until telomeres become too short to perform their role and preserve their ability to divide. It results in cellular aging being one of the main causes of aging of the whole body. According to the results of the present study including samples even of similar age (18–25 years), we detected a significant negative effect of age on shortening of telomeres.

Table 2. Multiple regression analysis demonstrating the effect of *TERC* rs7726159, *TERC* rs1317082, and *OBFC1* rs2487999 gene variants and sociodemographic factors (age and place of residence) on individual differences in telomere length and depression level

DV	Predictor	Group	Model 1		Model 2		Model 3		Model 4		
			β	p-value	β	p-value	β	p-value	β	p-value	
RTL	Intercept		0.949	<2e-16	0.990	<2e-16	0.997	<2e-16	0.96	<2e-16	
	<i>TERC</i> rs7726159	A/C	0.076	0.015	–	–	–	–	0.075	0.017	
		A/A	0.044	0.373	–	–	–	–	0.043	0.389	
	<i>TERC</i> rs1317082	A/G	–	–	0.003	0.917	–	–	–0.001	0.961	
		G/G	–	–	–0.019	0.687	–	–	–0.022	0.649	
	<i>OBFC1</i> rs2487999	C/T	–	–	–	–	–0.042	0.302	–0.038	0.347	
		T/T	–	–	–	–	–0.163	0.404	–0.167	0.392	
	Model p-value		0.052		0.893		0.426		0.265		
	Corrected r^2		3.8e-3		<2e-16		<2e-16		1.6e-3		
		Intercept		1.53	<2e-16	1.483	<2e-16	0.918	<2e-16	1.478	<2e-16
RTL	<i>TERC</i> rs7726159	A/C	–	–	0.074	0.020	0.080	0.017	0.078	0.019	
		A/A	–	–	0.039	0.439	0.064	0.239	0.051	0.348	
	Age		–0.027	1e-3	–0.026	0.001	–	–	–0.027	0.001	
	Place of residence	Rural	0.059	0.066	–	–	0.069	0.032	0.063	0.048	
	Model p-value		5.9e-4		9.2e-4		0.019		4.3e-4		
	Corrected r^2		0.015		0.013		0.008		0.018		
		Intercept		9.261	<2e-16	9.120	<2e-16	8.867	<2e-16	9.145	<2e-16
	RTL		–0.643	0.206	–0.661	0.155	–0.643	0.166	–0.605	0.195	
	<i>TERC</i> rs7726159	A/C	–0.601	0.239	–	–	–	–	–0.530	0.263	
		A/A	–0.274	0.739	–	–	–	–	–0.176	0.814	
Depression	<i>TERC</i> rs1317082	A/G	–	–	–0.139	0.768	–	–	–0.073	0.877	
		G/G	–	–	–0.216	0.769	–	–	–0.149	0.840	
	<i>OBFC1</i> rs2487999	C/T	–	–	–	–	0.961	0.110	0.931	0.123	
		T/T	–	–	–	–	–1.664	0.568	–1.605	0.583	
	Model p-value		0.356		0.542		0.174		0.505		
	Corrected r^2		2.7e-4		<2e-16		0.002		<2e-16		

DV—dependent variable; RTL—relative telomere length; β —regression coefficient. The reference groups in the models were for *TERC* rs7726159, *TERC* rs1317082, *OBFC1* rs2487999 minor genotypes and for place of residence urban residency. Statistically significant differences are shown in bold.

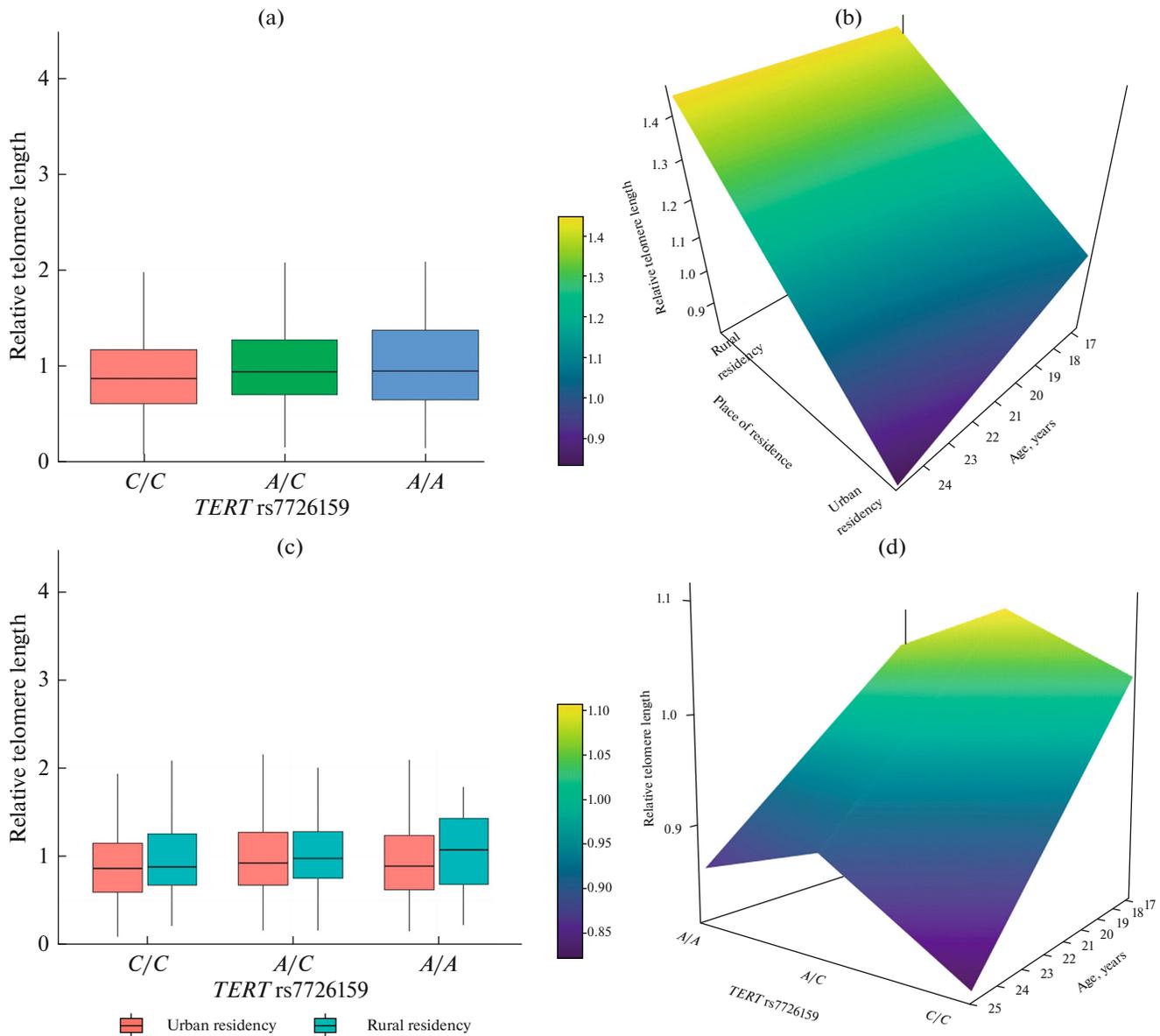


Fig. 1. Association of relative telomere length and allelic variants of *TERT* rs7726159 (a), age and place of residence (urban/rural residency) (b), mentioned genetic and environmental predictors simultaneously (c, d).

Together with the influence of age, in the present study, we revealed a negative effect of urbanization on shortening of telomeres and the main effect of rs7726159 in the *TERT* gene. A negative effect of urban residency in childhood is attributed to the fact that this period of ontogenetic development is the most sensitive to adverse environmental factors, which is related to the anatomical and physiological characteristics of the child's body, such as low efficiency of barrier factors of the upper respiratory tract, increased permeability of the blood-brain barrier and mucous membranes, limited excretory kidney function, and growth-dependent enlargement in the fat and bone mass tissues, and contributes to the deposition of toxic

substances. At the same time, the immaturity of enzymatic systems determines a reduced efficacy of local immunity factors, antioxidant defense, and detoxification system of chemical substances [25]. Previously, our research group also demonstrated an unfavorable effect of rearing in urban residency on human cognitive abilities in the context of gene-by-environmental interactions, which was significant even in the present genetically "favorable" allele in the apolipoprotein E (*APoE*) gene [26]. A large number of published studies indicate a statistically significant influence of an unfavorable environment on shortening of telomeres, which, according to one of the studies [18], corresponds to a decrease in chronological age by 8.7–11.9 years in

numerical equivalent (or 69–174 bp) with worsening environmental conditions. A negative impact of living in a high-tech society compared to small territorial units is primarily caused by the annual emission of millions of tons of toxic substances into the atmosphere by industry and as exhaust gases. It is known that heavy metals (for example, manganese), whose concentration is exceeded in urban locations compared to rural areas [25], have a negative impact on the central nervous system, resulting in sensitization of the body and the development of neurodegenerative diseases [27]. Animal studies demonstrated the effect of welding smoke on changes in telomere length in brain tissues, which is associated with the regulation of the shelterin complex components (Trf1 and Trf2) without modifications in telomerase activity, on one hand, and enhanced level of biomarkers associated with neurodegenerative changes, on the other hand [27].

According to the large-scale study based on summary information on concentrations of different 37 xenobiotics obtained from the Registry of the National Air Toxics Assessment, the exceeding of the level of several ecotoxicants in the atmosphere (benzidine, 1,4-dioxane, carbon tetrachloride, chloroprene, ethylene dibromide, and propylene dichloride) is associated with shortening of telomeres caused by initiation of the processes of oxidative stress in the body [28]. A negative impact of urbanization and related environmental pollution has been demonstrated on both human body and other living objects. For instance, the study of a link between the concentration of several xenobiotics (phthalates, organochlorine and pyrethroid pesticides, polychlorobiphenyls, polybromodiphenyl esters, polycyclic aromatic hydrocarbons, and their metabolites) and telomere length in European chub (*Squalius cephalus*) reported a negative correlation between the level of phthalate metabolites and the length of telomere repeats, as well as dwelling in the hydrographic network of urban reservoirs [29].

It should be mentioned that air pollution has an adverse effect on cellular aging of the body (reducing telomere length) even within prenatal development [30]. Since the place of residency (urban/rural) in childhood to a large extent correlates with the maternal place of residence during gestation, a significant adverse effect of prenatal development under urban conditions on shortening of telomeres may be caused by molecular events in the placenta and related to the presence of toxic substances in the environment. In particular, small dispersed particles PM_{2.5} (less than 2.5 μm in size) can easily penetrate through the hemotoplacental barrier and affect fetal development owing to the formation of active oxygen and nitrogen forms. Small dispersed particles may possess free radicals on their surface and form active hydroxyl radicals during the Fenton reaction in the presence of transition metals (e.g., iron) on the particle surface [31]. On the other hand, generation of active oxygen forms may be related to the changes in functioning of NADPH oxi-

dases induced by solid particles and activation of inflammatory cells [30]. Fine aerosol influences the future organism via epigenetic and genetic modifications. In particular, the global level of DNA methylation (LINE-1 mobile elements) in the placenta negatively correlates with the effect of small dispersed particles on the body of the mother and fetus [30]. In turn, a decrease in global level of DNA methylation has an adverse influence on telomere length [32]. Moreover, the effect of PM_{2.5} particles may modify expression of microRNAs in the placenta and in certain regions of the developing brain [33], which points to a significant role of these modifications in neurobiological processes.

One of the accompanying manifestations of urban residency is the impact of anthropogenic noise on a living organism, which can be especially crucial at early stages of development. Noise can represent a stress factor and result in cellular aging, which is expressed as shortening of telomeres, for instance, in great tits (*Parus major*) nesting in urban versus rural locations [34]. On the other hand, several authors also reported a link between green territory in a neighborhood and longer telomeres [35]. Probably, individuals living in unfavorable conditions possess significant allostatic load (aging of all systems of the body) and chronic activation of stress response, which results in hyperfunctioning of the HPA system and excess of stress hormones, thus contributing to shortening of telomeres.

At the molecular level, the impact of environmental factors in regulation of the length of telomeres can be attributed to their effect causing changes in the epigenetic profile. Multiple findings associated the level of DNA methylation with a negative effect of pollutants (see review [36]). At the same time, a safe environment has positive consequences on enhanced length of telomeres owing to hypomethylation. In particular, meditative practices result in a drop in methylation of the region located on a short arm of chromosome 4, which is characterized by the absence of protein-encoding genes and neighboring insulator region [37]. DNA methylation in subtelomeric regions is assumed to play a significant role in maintaining the length of telomeres [37]. For instance, TET (ten-eleven translocation, methylcytosine dioxygenase), which is responsible for 5-methylcytosine conversion to 5-hydroxymethylcytosine, may be crucial for the maintenance of the structure of telomeres owing to modulation of DNA methylation in subtelomeric regions [38]. A significant role of DNA methylation in telomere homeostasis has been confirmed by animal studies, which reported diminished global methylation level in subtelomeric regions providing excessive elongation of telomeres in murine embryonic stem cells with “switched-off” DNA methyltransferases [39]. Moreover, it is assumed that stress effects on the epigenome are caused by stress-sensitive transposons, which initiate the process of formation of noncoding

RNAs affecting modification of histones and methylation of certain genetic loci [40].

Therefore, as a result of the present study, a large dataset including the information on relative telomere length, variants of genes encoding the proteins involved in the regulation of telomere length, and sociodemographic parameters has been analyzed for the first time in mentally healthy individuals aged 18–25 years. We confirmed the impact of genetic variant rs7726159 in the reverse transcriptase gene (*TERT*) and age in regulation of telomere length in the examined cohort. Moreover, a negative effect of rearing in urban residency in childhood on shortening of telomeres was observed, which characterizes the level of cellular aging of the body. Despite the suggested hypothesis on a relation of depression level (not involving clinical forms) to decreased length of telomeres, we failed to detect such a relation in the present study in young adults. The present study is characterized by several advantages, including a large sample size homogenous by age and collected prior the COVID-19 pandemic, which makes it possible to assess a psychoemotional state prior the effect of SARS-CoV-2, which is known to negatively affect human health. Another advantage of the present research includes an attempt to associate the specificity of childhood rearing to the changes in telomere length later in adulthood. Nevertheless, we did not account for several important components of a gene network involved in regulation of telomere length and adverse stress effects early in ontogenesis, which also contribute to a shortening of chromosomal telomeres.

FUNDING

The present study was supported by the Russian Science Foundation (project no. 17-78-30028) (in the part of psychological testing and collection of biological samples), megagrant of the Ministry of Science and Higher Education of the Republic of Bashkortostan in the part of genetic analysis, and megagrant of the Ministry of Science and Higher Education of Russian Federation (project no. 075-15-2021-595) in the part of statistical and bioinformatic analysis.

COMPLIANCE WITH ETHICAL STANDARDS

Conflict of interest. The authors declare no conflict of interest.

Statement of compliance with standards of research involving humans as subjects. All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki Declaration and its later amendments or comparable ethical standards. Informed consent was obtained from all individual participants involved in the study.

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Translated by A. Kazantseva