Research Square

Preprints are preliminary reports that have not undergone peer review.
They should not be considered conclusive, used to inform clinical practice,
or referenced by the media as validated information.

Sociocultural stress and SLC6A4 gene polymorphism contribute to telomere shortening in depression

Hana Shimizu-Furusawa
hana-shimizu@umin.ac.jp

Teikyo University
Handayani Halik
Hasanuddin University
Andi Agus Mumang
Hasanuddin University
Kristian Liaury
Hasanuddin University
Irawan Yusuf
Hasanuddin University
Andi Jayalangkara Tanra
Hasanuddin University
Kei Asayama
Teikyo University
Takayoshi Ohkubo
Teikyo University
Izumi Naka
The University of Tokyo
Jun Ohashi
The University of Tokyo
Takafumi Ishida
The University of Tokyo
Takuro Furusawa
The University of Tokyo

Article

Keywords: telomere, depression, stress, serotonin transporter, SLC6A4 genotype, case-control study

Posted Date: November 3rd, 2026

DOI: https://doi.org/10.21203/rs.3.rs-11303264/v1

License: This work is licensed under a Creative Commons Attribution 4.0 International License. Read Full License

Additional Declarations: No competing interests reported.

Abstract

Telomeres shorten due to environmental and psychological stress. Psychiatric disorders, especially those triggered by stress, can alter telomere length. Genetic factors associated with depression vary across cultures and environments. The serotonin-transporter-linked polymorphic region (5-HTTLPR) of *SLC6A4* has been associated with susceptibility to depression, but the risk allele appears to differ between populations. This study aimed to investigate the correlation between telomere length and depression in Indonesia, focusing on socioeconomic, cultural, and genetic factors amidst rapid social changes. We recruited 97 psychiatric outpatients and 101 participants without depressive symptoms. Psychological assessments were conducted using the Self-Construal Scale (SCS) and Behavioral Inhibition System/Behavioral Activation System (BIS/BAS) via interviews. DNA extracted from the peripheral blood was used to identify the *SLC6A4* genotype by PCR, and telomere length was measured by quantitative PCR. The comparison of patients with depression and controls revealed significant telomere shortening in the patient group ($p < 0.0001$, Mann-Whitney U-test). Logistic regression showed that psychological scales were stronger predictors of depression than telomere length or *SLC6A4* polymorphisms. A multiple regression analysis indicated that the LL genotype of *SLC6A4*, but not socioeconomic or psychological factors, was strongly associated with telomere length ($p = 0.001$). This study is the first to analyze both psychological and genetic factors in the relationship between depression and telomere shortening, providing new insights into the genetic co-evolution mechanisms associated with depression.

INTRODUCTION

Telomeres play a crucial role in cellular aging as they shorten with each cell division; however, this shortening has been linked to various environmental and psychological stresses and lifestyle factors that can accelerate the aging process [1,2]. Research has demonstrated that psychiatric disorders, particularly those triggered by life stressors, can lead to changes in telomere length. For example, a systematic review compiled evidence from several studies that measured telomere length in individuals experiencing anxiety, depression, or related disorders and suggested that psychiatric conditions might have contributed to telomere shortening [3]. Additionally, Ellaway, et al. [4] found an association between higher levels of neighborhood stress and shorter telomeres. Notably, Gillis, et al. [5] provided evidence that longer telomere length at midlife is associated with a reduced risk of worsening depressive symptoms over a span of 20 years, particularly among women.

In contemporary society, the significance of mental and social well-being is increasingly being acknowledged alongside physical health. Stress-related mental health issues, particularly depression, have emerged as major global challenges [6]. Numerous studies have shown that factors contributing to depression include genetic and biological components, lifestyle, and environmental factors [7,8]. The relationship between telomere length and depression has recently garnered attention. It is possible that the development of depression could affect telomere length or that a common factor, such as stress, could influence both. Nevertheless, the Netherlands Study of Depression and Anxiety (NESDA) revealed that while polygenic risk scores (PRS) for depression were correlated with lifetime depression and its severity, they did not show a relationship with telomere length [9]. However, research on the complex relationship between these two factors is limited.

The risk factors and causal mechanisms of depression can vary among countries and ethnic groups living in different cultures and environments [10,11]. In addition, genetic epidemiological studies suggest that polymorphisms in the serotonin transporter gene solute carrier family 6 member 4 (*SLC6A4*) contribute to susceptibility to psychiatric disorders differently in European and East Asian communities [7,10]. Despite a growing body of research, most studies on telomeres have been conducted in developed countries. This underscores the urgent need to investigate these relationships in developing nations, which potentially display diversity in the biological and social mechanisms associated with cellular aging and mental health. These nations have also been facing challenges related to the aging of society and increased mental health issues [6].

The prevalence of depression has been rapidly increasing in Indonesia [12], the largest country in Southeast Asia with a population of 260 million. However, only one study has explored the relationship between job stress and telomere length among nurses in Bali, reporting a negative correlation [13]. This study aimed to investigate the correlation between telomere length and depression in Indonesia during a time of rapid social and cultural changes. Special attention was paid to exploring the socioeconomic, cultural, and genetic factors for depression and telomere length.

METHODS

Recruitment of the participants

This was a case-control study conducted in Makassar, the capital city in the province of South Sulawesi, Indonesia. Outpatients at the Department of Psychiatry, Hasanuddin University, underwent general clinical checks for the diagnosis of depression by licensed Indonesian psychiatrists. Once diagnosed with depression based on the Indonesian Mental Disorder Guidelines for Classification and Diagnosis (*Pedoman Penggolongan dan Diagnosa Gangguan jiwa edisi 3*), patients who agreed to participate in this study underwent further questionnaires and blood sampling.

Healthy adult volunteers without any medication were recruited as controls by an investigator (AAM) visiting three local residential areas in Makassar. Psychiatrists confirmed that all the control participants showed no depressive symptoms when they visited the affiliated hospital for blood sampling. Detailed procedures for the interviews and collection of biological specimens have been described elsewhere [8]. In total, 198 participants (97 cases and 101 controls) underwent an examination of telomere length and genotyping using DNA extracted from whole blood. Several participants were excluded from the following statistical analysis due to missing data for each acquired parameters.

All participants provided their written informed consent prior to enrollment. This research was approved by the Ministry of Education and Culture Ethics Committee of Medical Research Indonesia (approval number: 01/H4.8.4.5.31/PP36-KOMETIK/2017) to the Faculty of Medicine, Hasanuddin University and

Hasanuddin University Hospital, Kyoto University Graduate School and Faculty of Medicine, Ethics Committee (approval number: G1099) to the Graduate School of Asian and African Area Studies, Kyoto University, and by the School of Medicine, Teikyo University Ethics Committee (approval number: 22–190) to the Department of Hygiene and Public Health, School of Medicine, Teikyo University. All methods were performed in accordance with the relevant guidelines and regulations.

Questionnaire and blood collection

Face-to-face interviews using demographic and socioeconomic questionnaires and cultural psychological scales were conducted. The first part of the questionnaire included questions on age, sex, religion, education, and occupation. The second part measured the cultural psychological attitudes of each participant, and the Singelis Self-Construal Scale (SCS) was used to evaluate independent and interdependent self-construal [14] and the Behavioral Inhibition System and Behavioral Activation System (BIS/BAS) were used to assess motivation focus [15]. The development and validity of the Indonesian versions of these common scales have been described elsewhere [16].

Diagnoses and medical records regarding the onset of symptoms and prescribed drugs were collected by the psychiatrists in charge. Venous blood was collected at each hospital, transported to Hasanuddin University Medical Research Center in Makassar, and kept frozen at − 30°C until DNA extraction.

DNA extraction and genetic analyses

Genomic DNA was extracted from 200 µL of whole blood using a commercial DNA extraction kit (QIAamp DNA Blood Mini Kit, Qiagen, Netherlands) based on silica gel DNA-binding membrane spin columns. An aliquot of DNA was used for the genotyping and quantification of telomere length. The DNA concentration was measured using a spectrophotometer (GENESYS 10 UV, Thermo Electron Corporation, USA). *SLC6A4* encodes the serotonin transporter, which mediates the reuptake of serotonin from the synaptic cleft and thereby regulates serotonergic neurotransmission. The most widely studied functional variant of this gene is 5-HTTLPR, a length polymorphism in the 5' promoter region that consists of a variable number of GC-rich repeat units of 20–23 bp. The insertion/deletion of a 43-bp segment produces a long (L) allele and a short (S) allele, and the L allele is associated with higher transcriptional activity and serotonin uptake than the S allele. The S allele has been reported as a risk factor for depression under life stress in a European-ancestry cohort[7], whereas the L allele has been associated with anxiety in Han Chinese[11]. It has been proposed that this difference reflects culture–gene coevolution, with the S allele being more frequent in collectivistic East Asian societies[10].

The 5-HTTLPR genotype was determined using polymerase chain reaction (PCR). The forward primer for the 5-HTTLPR gene was [5′-GCG CTC CTG CAT CCC CCA TTA-3′] while the reverse primer used was [5′-GGG ATG CGG GGG AAT ACT GGT-3′]. The PCR reaction was carried out in a total volume of 10 µL, which included 4.1 µL of PCR-grade water, 5 µL of AmpliTaq Gold 360 Master Mix, 0.2 µL of each primer (forward and reverse), and 0.5 µL of DNA template. The thermal cycling protocol was as follows: initial denaturation at 95°C for 10 min, followed by 25 cycles of denaturation at 95°C for 30 s, annealing at 65°C for 35 s, and extension at 72°C for 30 s, with a final extension at 72°C for 7 min. PCR products were separated by electrophoresis on a 2% agarose gel.

Measurement of telomere length

Telomere length of peripheral blood mononuclear cells was determined from extracted DNA using a real-time quantitative PCR method described elsewhere [2], which was originally developed by Cawthon [17] and modified [18]. Two independent PCRs were run to determine the ratio of telomere repeat copy number (T) to single copy gene number (S) as a T/S ratio for each sample using the ΔΔCT method. Quantitative PCR was carried out in a 20 µL reaction volume using 5 ng template DNA and Light Cycler FastStart DNA Master SYBR Green I (Roche Diagnostics K.K., Tokyo, Japan). The primers for telomere PCR were tel1b [5′-CGG TTT GTT TGG GTT TGG GTT TGG GTT TGG GTT TGG GTT TGG GTT-3′] at a final concentration of 100 nM and tel2b [5′-GGC TTG CCT TAC CCT TAC CCT TAC CCT TAC CCT-3′] at a final concentration of 900 nM. The primers for the single-copy gene 36B4 were 36B4u [5′-CAG CAA GTG GGA AGG TGT AAT CC-3′] at a final concentration of 300 nM and 36B4d [5′-CCC ATT CTA TCA TCA ACG GGT ACA A-3′] at a final concentration of 500 nM. PCRs were run on a Light Cycler 2.0 Instrument (Roche Diagnostics K.K., Tokyo, Japan). Thermal cycling conditions were 40 cycles of denaturation at 96°C for 1 s and anneal/extension at 54°C for 60 s for telomeres, and 40 cycles of denaturation at 95°C for 15 s and annealing at 58°C for 60 s for 36B4. Each PCR run included the same reference DNA pooled from all individuals for quality control, and melting curve analysis was performed to confirm the specificity of each amplification. All samples were amplified in duplicate. The cycle threshold (Ct) of telomeres or 36B4 was converted to ΔΔCt. Telomere length was determined as the T/S ratio ($2^{-\Delta\Delta Ct}$) of each sample relative to the average T/S of the control samples using the following formula: $[2^{Ct(telomere)} / 2^{Ct(36B4)}]^{-1} = 2^{-\Delta Ct}$; $2^{-(\Delta Ct(each\ sample) - \Delta Ct(mean\ of\ control\ samples))} = 2^{-\Delta\Delta Ct}$.

Statistical analysis

All statistical analyses were conducted using R (ver. 4.1.3). The Mann-Whitney U-test and Fisher's exact test were used for non-parametric comparison of mean values and proportions between two groups, respectively, and Kruskal-Wallis test for 3-group comparison. The correlation coefficient was calculated using Spearman's rank test.

This study compared four models to detect the association between telomere length and the occurrence of depression by adjusting for different sets of exploratory variables. Model 1 was the fundamental model and included age, sex, and socioeconomic status (educational history and occupation), which are known to be elevated risk factors for the occurrence of depression [8]. Model 2 included Singelis' self-construal scales and the BIS/BAS scales, which are also known to be associated with the occurrence of depression in Indonesia [16] in addition to all variables in Model 1. Model 3 included polymorphisms in *SLC6A4*

(two dummy variables: LL = 1 and LS = 1, while SS = 0 for both), which had also been a major genetic factor for depression [7], in addition to all variables in Model 1. Model 4 included all variables (i.e., age, sex, socioeconomic status, self-construal scales, BIS/BAS scales, and *SLC6A4* polymorphism). We assumed these models because there are two hypotheses that variables affecting depression and telomere length are identical [19] or different [9]. We ran four models for the logistic regression analysis with the occurrence of depression as the dependent variable and for the general linear model (GLM) with relative telomere length as the dependent variable. The Akaike Information Criteria (AIC) were calculated to select the final regression model.

To confirm the association between the dependent and explanatory variables, GLM was run for the case and control, respectively, based on the best-fitted model. Finally, an analysis of covariance (ANCOVA) based on the best-fitted model was run to clarify the effects of genotype and depression on telomere length, and Tukey's honestly significant difference (HSD) test was applied for multiple comparisons among different groups. Statistical significance was set at $P < 0.05$, and a smaller AIC was considered a better model.

Data Availability

The data supporting the findings of this study are available from the corresponding author upon reasonable request, subject to ethical approval and applicable data protection requirements. Individual-level genetic and clinical data are not publicly available because of privacy and confidentiality considerations.

RESULTS

Table 1 presents the characteristics of the participants. There were no significant differences in sex or age between the case and the control groups. The majority of participants were Muslim (92% in the control group and 89% in the case group). Regarding educational background, 72% of the case group had received high school education or higher, compared to 52% of the control group. While employment in the private sector was common among the case, no regular job (no employment) was more prevalent in the control group. The case group was less independent in self-construal (5.09 ± 0.62 vs. 5.58 ± 0.85, $P < 0.0001$), and had lower BAS Drive and Fun Seeking scores (11.09 ± 2.70 vs. 13.28 ± 2.15, $P < 0.0001$) and lower BAS Reward Responsiveness scores (14.13 ± 3.74 vs. 17.81 ± 2.39, $P < 0.0001$) compared to the control group. There were no differences in the allele frequencies at the *SLC6A4* locus. The relative telomere length was shorter in the case group (− 2.26 ± 4.55) than in the control group (0.00 ± 3.83).

Table 1
Characteristics of the participants

Age (year) mean ± SD	**Control (n = 101)**			**Case (n = 97)**			***P***
	42.61	**±**	**12.23**	**45.98**	**±**	**13.39**	**NS**
Sex; male (female)	33		(68)	46		(53)	NS
Education							0.011
Postgraduate	4		(4%)	2		(2%)	
Bachelor	24		(24%)	26		(26%)	
Diploma	4		(4%)	7		(7%)	
High school	20		(20%)	37		(37%)	
Junior high school	19		(19%)	16		(16%)	
Elementary school	27		(27%)	10		(10%)	
Drop out of elementary school	3		(3%)	1		(1%)	
Religion							NS
Islam	93		(92%)	88		(89%)	
Christian	4		(4%)	9		(9%)	
Others	4		(4%)	2		(2%)	
Occupation							0.003
Civil servant	14		(14%)	10		(11%)	
Private business	11		(11%)	39		(41%)	
Housewife	25		(25%)	25		(26%)	
Retired	2		(2%)	7		(7%)	
No regular job	49		(49%)	14		(15%)	
Self-Construal Scale (SCS) for Indonesians							
SCS independent	5.58	±	0.85	5.09	±	0.63	< 0.0001
SCS interdependent	4.20	±	1.13	4.34	±	0.91	NS
Behavioral Inhibition System (BIS) / Behavioral Activation System (BAS) for Indonesians							
BIS	11.54	±	2.76	10.99	±	3.18	NS
BAS Drive + Fun seeking	13.28	±	2.15	11.09	±	2.70	< 0.0001
BAS Reward responsiveness	17.81	±	2.39	14.13	±	3.75	< 0.0001
Genotype *SLC6A4* [a]							NS
LL	21		(21%)	10		(11%)	
LS	36		(36%)	39		(43%)	
SS	43		(43%)	42		(46%)	
Relative telomere length to control							
− ΔΔCt	0.00	±	3.83	−2.26	±	4.55	< 0.0001

[a] Serotonin-transporter-linked polymorphic region (5-HTTLPR) in the promoter of SLC6A4 (solute carrier family 6 member 4). L, long allele; S, short allele.

The independent self-construal scale was correlated with relative telomere length when both the case and control groups were combined (ρ = 0.20, *P* = 0.0056). However, when the case and control groups were analyzed separately, neither the independent self-construal scale nor the interdependent self-construal scale showed any correlation (Fig. 1). When the case and control groups were combined, BIS (ρ = 0.26, *P* = 0.0003), BAS Drive and Fun Seeking (ρ = 0.23, *P* = 0.0013), and BAS Reward Responsiveness (ρ = 0.31, *P* < 0.0001) were correlated with telomere length. However, when the groups were analyzed separately, the case group showed correlations between telomere length and BIS (ρ = 0.30, *P* = 0.0030), BAS Drive and Fun Seeking (ρ = 0.25, *P* = 0.0127), and BAS Reward Responsiveness (ρ = 0.35, *P* = 0.0004), whereas in the control group, only BIS was correlated with telomere length (ρ = 0.22, *P* = 0.0333) (Fig. 2).

Table 2 shows association between treatment and the relative telomere length. Within the case group, the majority of the case group were prescribed for dual therapy and displayed the shorter telomere length than a group with no medicinal treatment (*P* = 0.004). In addition, the case treated with Clobazam

showed significantly shorter telomere length than the group with no medicinal treatment ($P = 0.04$); it should be noted that 13 out of 15 patients who were treated with Clobazam were assigned to dual therapy. Although not shown in the figure, the duration of being diagnosed as depression was not associated with the relative telomere length in the case ($\rho = 0.14$, NS).

Table 2
Antidepressant drug prescription and the relative telomere length

	Classification of drugs			Case (n = 97)					
	ATC code	**ATC pharmacological subgroup**	**ATC chemical subgroup**	**N**	**(%)**	**Relative telomere length (-ΔΔCt)**			***P***
Prescribed drugs									
None	—	—	—	8	(8%)	1.76	±	0.35	referent
Fluoxetine	N06AB03	ANTIDEPRESSANTS	Selective serotonin reuptake inhibitors	72	(74%)	-2.56	±	4.50	NS
Alprazolam	N05BA12	ANXIOLYTICS	Benzodiazepine derivatives	49	(51%)	-2.61	±	4.78	NS
Clobazam	N05BA09	ANXIOLYTICS	Benzodiazepine derivatives	15	(15%)	-3.98	±	4.33	0.039
Risperidone	N05AX08	ANTIPSYCHOTICS	Other antipsychotics	9	(9%)	-2.64	±	2.84	NS
Olanzapine	N05AH03	ANTIPSYCHOTICS	Diazepines, oxazepines, thiazepines and oxepines	7	(7%)	0.21	±	3.77	NS
Trihexyphenidyl	N04AA01	ANTICHOLINERGIC AGENTS	Tertiary amines	7	(7%)	-1.60	±	3.71	NS
Number of prescriptions									
None				8	(8%)	1.76	±	0.35	referent
1				9	(9%)	0.21	±	2.79	NS
2				60	(62%)	-3.65	±	4.77	< 0.001
≥ 3				20	(21%)	-0.44	±	2.86	NS

Table 3 compares the four models to identify the factors associated with the occurrence of depression. Across all the models, age, higher education and employment in the private sector were linked to an increased risk of depression. Additionally, independent and interdependent self-construal as well as BIS and BAS Reward Responsiveness were predictors of depression. The effect of telomere length was observed in Models 1 and 3, but disappeared in Models 2 and 4. The best model with the lowest AIC was Model 2, which included demographic, socioeconomic, cultural, and psychological variables but not genetic ones.

Table 3
Predictors of depression in 4 generalized linear models (logistic regression analyses)

Variables	Model 1				Model 2				Model 3				Model 4		
	OR	95%CI 2.5–97.5%		*P*	OR	95%CI 2.5–97.5%		*P*	OR	95%CI 2.5–97.5%		*P*	OR	95%CI 2.5–97.5%	
Relative telomere length	0.88	0.80	0.96	0.0043	0.93	0.83	1.04	NS	0.86	0.78	0.94	0.0017	0.91	0.81	1.02
Age (years)	1.04	1.02	1.07	0.0027	1.04	1.00	1.08	0.0316	1.04	1.01	1.07	0.0039	1.04	1.00	1.08
Sex (female = 1)	1.06	0.50	2.23	NS	1.64	0.63	4.45	NS	1.07	0.50	2.27	NS	1.65	0.61	4.60
Education: High	6.69	1.99	24.64	0.0028	8.11	1.68	46.27	0.0125	6.14	1.81	22.82	0.0048	7.72	1.56	44.86
Education: Middle	8.28	2.97	26.02	0.0001	12.23	3.25	55.51	0.0005	7.94	2.82	25.19	0.0002	12.05	3.17	55.72
Occupation: Public Sector	0.97	0.30	3.09	NS	0.87	0.20	3.71	NS	0.90	0.27	2.92	NS	0.79	0.17	3.47
Occupation: Private Sector	6.69	2.77	17.78	< 0.0001	5.13	1.72	16.82	0.0045	6.56	2.69	17.54	< 0.0001	5.01	1.66	16.75
Independent Self-construal					0.27	0.12	0.56	0.0008					0.27	0.12	0.55
Interdependent Self-construal					2.08	1.25	3.60	0.0061					1.98	1.19	3.44
BIS					1.24	1.04	1.51	0.0222					1.23	1.04	1.50
BAS Drive + Fun					0.95	0.71	1.26	NS					0.98	0.74	1.31
BAS Reward					0.70	0.55	0.87	0.0017					0.69	0.55	0.86
SLC6A4: LS									0.85	0.40	1.81	NS	1.03	0.40	2.67
SLC6A4: LL									0.38	0.12	1.10	NS	0.42	0.10	1.53
(Intercept)	0.01	0.00	0.09	< 0.0001	17.16	0.29	1272.20	NS	0.02	0.00	0.13	< 0.0001	23.86	0.36	2032.30
McFadden pseudo R^2				0.202				0.429				0.215			
AIC [a]				212.07				166.15				212.84			

[a] Akaike Information Criterion

Table 4 shows four models for detecting factors of the telomere length. Interestingly, known factors for the depression (i.e., age, sex, socioeconomic status, and cultural psychological scales) were not detected in these models (except for BIS in Model 2). Inversely, the LL genotype of the *SLC6A4* was the significant factor for Models 3 and 4. Depression (the case) was a significant factor in Models 1 and 3, while the best model was Model 4. When the Model 4 (excluding the variable "case") was run separately for the case and the control, the LL genotype was the only significant factor in the case group ($P = 0.00342$, model adjusted $R^2 = 0.1316$), and no appropriate model was found for the control group. When dual therapy or the use of clobazam was included in the model for the case, the use of these medicine as well as the LL genotypes were the only significant variables (Table 5).

Table 4
Predictors of relative telomere length in 4 general linear models

Variables	Model 1			Model 2			Model 3			Model 4		
	β	**SE**	***P***	***β***	**SE**	***P***	***β***	**SE**	***P***	***β***	**SE**	***P***
Depression (Case = 1)	-2.07	0.70	0.003	-0.97	0.82	NS	-2.24	0.68	0.001	-1.22	0.80	NS
Age (years)	0.00	0.03	NS	0.01	0.02	NS	0.00	0.03	NS	0.01	0.02	NS
Sex (female = 1)	-0.62	0.69	NS	-0.94	0.69	NS	-0.57	0.68	NS	-0.82	0.67	NS
Education: High	-0.02	1.05	NS	0.19	1.04	NS	-0.15	1.03	NS	0.07	1.01	NS
Education: Middle	0.51	0.88	NS	0.25	0.87	NS	0.39	0.86	NS	0.13	0.85	NS
Occupation: Public Sector	0.02	1.11	NS	-0.03	1.08	NS	-0.22	1.08	NS	-0.25	1.05	NS
Occupation: Private Sector	0.22	0.81	NS	0.29	0.80	NS	0.17	0.79	NS	0.32	0.78	NS
Independent Self-construal				0.47	0.49	NS				0.21	0.48	NS
Interdependent Self-construal				-0.21	0.35	NS				-0.28	0.34	NS
BIS				0.24	0.12	0.050				0.23	0.12	NS
BAS Drive + Fun				-0.14	0.21	NS				-0.03	0.20	NS
BAS Reward				0.26	0.16	NS				0.22	0.16	NS
SLC6A4: LS							-0.23	0.66	NS	-0.35	0.65	NS
SLC6A4: LL							-2.80	0.88	0.002	-2.88	0.88	0.001
(Intercept)	0.49	1.60	NS	-6.67	2.94	0.025	1.40	1.60	NS	-4.71	2.94	NS
Adjusted R^2		0.030			0.087			0.078			0.135	
P		0.095			0.007			0.007			0.0005	
AIC [a]		1019.11			1013.02			1012.02			1005.25	

[a] Akaike Information Criterion

Table 5
Predictors of relative telomere length within the case group

Variables	*β*	SE	*P*
Age (years)	0.020	0.037	NS
Sex (female = 1)	-0.100	1.045	NS
Education: High	-0.623	1.724	NS
Education: Middle	-0.069	1.588	NS
Occupation: Public Sector	2.266	1.761	NS
Occupation: Private Sector	0.515	1.105	NS
Independent Self-construal	0.341	0.897	NS
Interdependent Self-construal	-0.562	0.547	NS
BIS	0.017	0.228	NS
BAS Drive + Fun	-0.002	0.326	NS
BAS Reward	0.310	0.238	NS
SLC6A4: LS	-0.252	0.983	NS
SLC6A4: LL	-3.562	1.633	0.033
Dual therapy or Clobazam use	-2.724	0.995	0.008
(Intercept)	-4.467	4.874	NS
Adjusted R^2		0.160	
P		0.02	
AIC [a]		468.48	
[a] Akaike Information Criterion			

Figure 3 shows the relative telomere length, broken down by genotype and case/control status, after adjusting for age, sex, education, occupation, and cultural psychological scales in an ANCOVA model. The results of the ANCOVA indicated that telomere length was the shortest in Case LL, followed by Case LS and Case SS. This was followed by the Control LL, Control LS, and Control SS, which had the longest telomere length. Tukey's HSD test revealed that Case LL had a significantly shorter telomere length relative to all of the other groups.

DISCUSSION

The key findings of this study are as follows: patients diagnosed with depression and undergoing treatment at a hospital in South Sulawesi, Indonesia exhibited apparently shorter telomere lengths than healthy citizens in the same location. This significant difference persisted even after adjusting for age, sex, and socioeconomic status, although it was not significant in models that included cultural, psychological, and/or genetic factors. While socioeconomic and psychological factors, as suggested by previous studies [8,16], remained influential on depression even after considering telomere length, the factors influencing telomere length were completely different. Specifically, polymorphisms in the serotonin transporter gene (*SLC6A4*) were associated with telomere length but not with depression or related factors. Notably, patients with long *SLC6A4* homozygotes had the shortest telomeres.

The association between telomere length and depression should be interpreted carefully because of the variety of factors and complexity of their relationships. Telomere length can change with age and sex because of its biological nature [20]; however, this study controlled for these major confounding factors by ensuring similar age and sex compositions in both the case and control groups. Additionally, this study found an association between psychiatric treatment (i.e., the use of clobazam or polypharmacy) and shortened telomere length, although it was difficult to establish causal relationships. Dual therapy was often prescribed when monotherapy failed to mitigate symptoms, while clobazam was used as an anxiolytic for severe symptoms. There are both possibilities that the treatment itself shortened the telomere length directly and that the effects of medicines within the case group might have indirectly reflected the severity of patients' conditions; further studies are necessary to identify its causal relationships. Even though the treatments directly affected telomere length, they did not affect the variables detected from the general linear models.

The important outcome of this study was the distinction between the risk factors for depression and telomere length. A meta-analysis of 38 studies (n = 34,347) on telomere length and depression showed a significant association, but the results of individual studies varied [21]. According to a GWAS, polygenic risk scores (PRS) for depression and telomere length showed very weak similarities [9]. This study supports those previous studies, as telomere length, but not depression, was associated with the *SLC6A4* genotype, particularly in patients with depression.

The short allele of *SLC6A4* has been recognized as a risk factor for depression in European populations [7]. Interestingly, the long allele is associated with anxiety and reduced connectivity between the prefrontal cortex and amygdala in Han Chinese [11]. A previous study suggested that this difference is due to

societal organization: individualism in the former and collectivism in the latter, possibly because genotype and cultural collectivism co-evolved [10]. Given that Indonesia, particularly the traditional society of South Sulawesi, is usually recognized as collectivistic, it is reasonable to assume that the long (LL) type became susceptible to stressors.

However, this study did not directly measure the stressors experienced by each participant. Further research is necessary to identify the stress factors that interact with *SLC6A4*, which leads to shortened telomere length and causal relationships with depression. Nonetheless, the study found that while telomere length was apparently associated with depression, its shortening was strongly determined by genetic factors rather than by environmental or cultural factors or depression itself in Indonesia. These findings provide new and important insights into the complex mechanisms related to cellular aging and mental disorders, and suggest the existence of mechanisms that vary among different ethnic groups. Future global research on telomere length and depression should carefully consider the influences of genetics, culture, and socioeconomics. Furthermore, the findings of this study could make a significant contribution to public health strategies in Indonesia, where an aging society is accompanied by an increase in mental health disorders.

CONCLUSIONS

A case-control study conducted in Indonesia revealed that (1) the telomere length was shortened in patients with depression; (2) the factors for telomere length were the LL genotype of the *SLC6A4* – a polymorphism at the serotonin transport gene and not those for depression (i.e., socioeconomic and cultural psychological factors); (3) the LL genotype of depression patients was associated with a significantly shorter telomere length in comparison to all other groups. Genetic and cultural factors that vary among ethnic groups influence the relationship between telomere length and depression, which has implications for addressing cellular aging in aging societies and mental health disorders amid rapid social changes.

Declarations

Authors' contributions

HSF, TF, JO, TI and YI conceptualized this study. HSF measured telomere length, analyzed the data, and wrote the first draft. HH carried out genotyping. AAM conducted sampling of the control participants. KL and AJT managed sampling of the patients in the hospitals. YI supervised sampling and laboratory analysis in Indonesia. KA offered suggestions on a draft. TO provided resources for writing. JO and IN determined the genotyping methods. TI was the principal investigator of the project. TF edited and finalized the manuscript. All authors read and approved the final manuscript.

Funding sources

This study was financially supported by Japan Society for the promotion of Sciences as KAKENHI Grant-in-Aid for Scientific Research (B) (16H05828, P.I. T. Ishida) and KAKENHI Grant-in-Aid for Scientific Research (C) (24K15467, P.I. H. Shimizu-Furusawa).

Interests' disclosure

The authors declared no conflict of interest.

Acknowledgements

We thank all the participants in this study. We also deeply thank the field contributors for control sampling, as well as the medical team of the Psychiatry Department at the Faculty of Medicine, Hasanuddin University, for recruiting case participants and conducting face-to-face interviews using questionnaires.

References

1. Lin, J. & Epel, E. Stress and telomere shortening: Insights from cellular mechanisms. *Ageing Res. Rev.* **73**, 101507. 10.1016/j.arr.2021.101507 (2022).
2. Fillman, T., Shimizu-Furusawa, H., Ng, C. F. S., Parajuli, R. P. & Watanabe, C. Association of cadmium and arsenic exposure with salivary telomere length in adolescents in Terai, Nepal. *Environ. Res.* **149**, 8–14. 10.1016/j.envres.2016.04.037 (2016).
3. Pousa, P. A. et al. Telomere shortening and psychiatric disorders: A systematic review. *Cells* **10**, 1423. 10.3390/cells10061423 (2021).
4. Ellaway, A., Dundas, R., Robertson, T. & Shiels, P. G. More miles on the clock: Neighbourhood stressors are associated with telomere length in a longitudinal study. *PLoS One*. **14**, e0214380. 10.1371/journal.pone.0214380 (2019).
5. Gillis, J. C. et al. The relation of telomere length at midlife to subsequent 20-year depression trajectories among women. *Depress. Anxiety*. **36**, 565–575. 10.1002/da.22892 (2019).
6. GBD 2021 Diseases and Injuries Collaborators. Global incidence, prevalence, years lived with disability (YLDs), disability-adjusted life-years (DALYs), and healthy life expectancy (HALE) for 371 diseases and injuries in 204 countries and territories and 811 subnational locations, 1990–2021: a systematic analysis for the Global Burden of Disease Study 2021. *Lancet* **403**, 2133–2161. 10.1016/S0140-6736(24)00757-8 (2024).
7. Caspi, A. et al. Influence of life stress on depression: moderation by a polymorphism in the 5-HTT gene. *Science* **301**, 386–389. 10.1126/science.1083968 (2003).
8. Mumang, A. A. et al. Socio-economic-demographic determinants of depression in Indonesia: A hospital-based study. *PLoS One*. **15**, e0244108. 10.1371/journal.pone.0244108 (2020).
9. Verhoeven, J. E., Penninx, B. W. J. H. & Milaneschi, Y. Unraveling the association between depression and telomere length using genomics. *Psychoneuroendocrinology* **102**, 121–127. 10.1016/j.psyneuen.2018.11.029 (2019).

10. Chiao, J. Y. & Blizinsky, K. D. Culture-gene coevolution of individualism-collectivism and the serotonin transporter gene. *Proc. Biol. Sci.* 277, 529–537, (2010). 10.1098/rspb.2009.1650
11. Long, H. et al. The long rather than the short allele of 5-HTTLPR predisposes Han Chinese to anxiety and reduced connectivity between prefrontal cortex and amygdala. *Neurosci. Bull.* **29**, 4–15. 10.1007/s12264-013-1299-x (2013).
12. Riset Kesehatan Dasar. *Laporan Nasional Riskesdas 2018*, < (2018). https://repository.badankebijakan.kemkes.go.id/id/eprint/3514/1/Laporan%20Riskesdas%202018%20Nasional.pdf
13. Purnawati, S., Dinata, I., Linawati, N. & Arijana, I. Correlation of telomere length and serotonin levels with job stress and locus of control type among nurses at a private hospital in Gianyar, Indonesia. *Acta Med. Philipp*. **55**, 632–639. 10.47895/amp.vi0.3160 (2021).
14. Singelis, T. M. The measurement of independent and interdependent self-construals. *Pers. Soc. Psychol. Bull.* **20**, 580–591. doi.org/10.1177/0146167294205014 (1994).
15. Carver, C. S. & White, T. L. Behavioral inhibition, behavioral activation, and affective responses to impending reward and punishment: The BIS/BAS Scales. *J. Pers. Soc. Psychol.* **67**, 319–333. https://doi.org/10.1037/0022-3514.67.2.319 (1994).
16. Istiqlal, T. et al. Self-construal and behavioral motivation systems among patients with depression in Indonesia: A hospital-based study. *Heliyon* **8**, e09839. 10.1016/j.heliyon.2022.e09839 (2022).
17. Cawthon, R. M. Telomere measurement by quantitative PCR. *Nucleic Acids Res.* **30**, e47. 10.1093/nar/30.10.e47 (2002).
18. Lin, J. et al. Analyses and comparisons of telomerase activity and telomere length in human T and B cells: Insights for epidemiology of telomere maintenance. *J. Immunol. Methods*. **352**, 71–80. 10.1016/j.jim.2009.09.012 (2010).
19. Vakonaki, E. et al. Common mental disorders and association with telomere length. *Biomed. Rep.* **8**, 111–116. 10.3892/br.2018.1040 (2018).
20. Rossiello, F., Jurk, D. & Passos, J. F. d'Adda di Fagagna, F. Telomere dysfunction in ageing and age-related diseases. *Nat. Cell. Biol.* **24**, 135–147. 10.1038/s41556-022-00842-x (2022).
21. Ridout, K. K., Ridout, S. J., Price, L. H., Sen, S. & Tyrka, A. R. Depression and telomere length: A meta-analysis. *J. Affect. Disord*. **191**, 237–247. 10.1016/j.jad.2015.11.052 (2016).

Figures

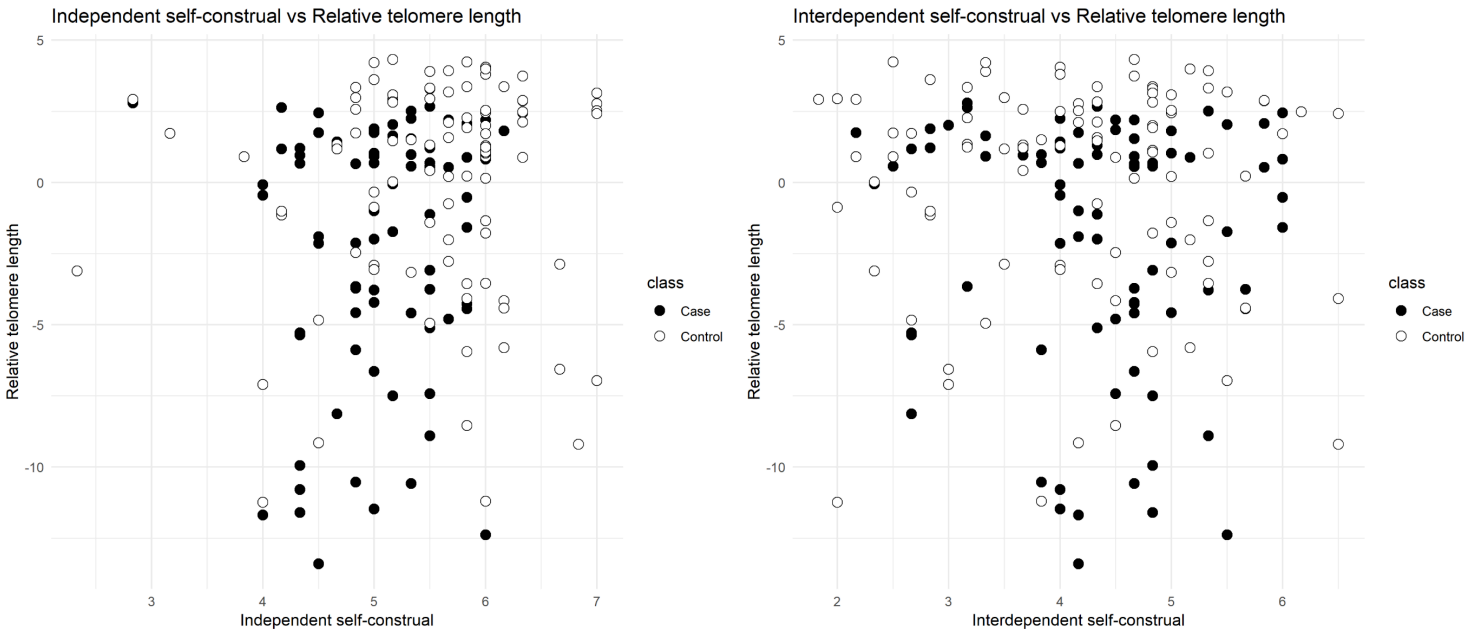


Figure 1

Relationship between relative telomere length and independent or interdependent self-construal scales

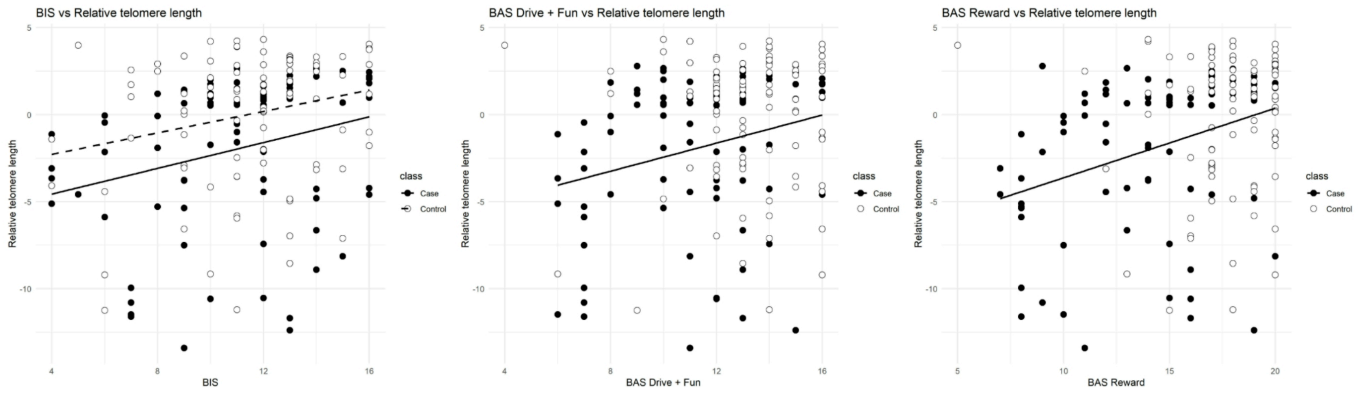


Figure 2

Relationship between relative telomere length and behavioral inhibition system (BIS) / behavioral activation system (BAS). Solid and dotted lines represent significant correlations in either case or control group.

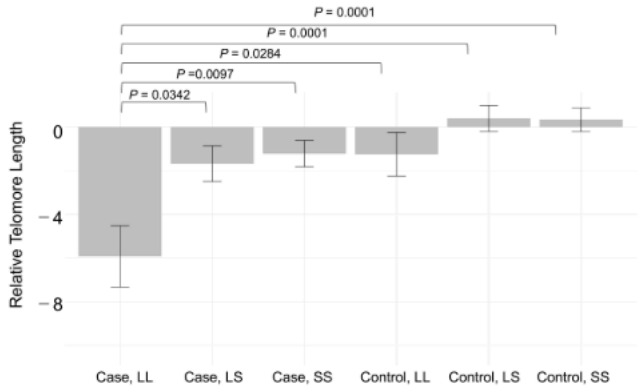


Figure 3

Relative telomere length broken down by *SLC6A4*genotype and the case/control group after adjusting for age, sex, education, occupation, self-construal scales, and BIS/BAS, with significant differences elucidated from an ANCOVA followed by Tukey's HSD test.