Heritability of telomere length in three generations of Korean families, including newborns

Jung-Ha Kim

Department of Medicine
The Graduate School, Yonsei University

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Directed by Professor Duk-Chul Lee

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Jung-Ha Kim

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of Jung-Ha Kim is approved.

Thesis Supervisor : Duk-Chul Lee

Thesis Committee Member#1 : Hye-Ree Lee

Thesis Committee Member#2 : Chung Mo Nam

Thesis Committee Member#3: Jaehong Ko

Thesis Committee Member#4: Jong Rak Choi

The Graduate School
Yonsei University

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ABSTRACT

Heritability of telomere length in three generations of Korean three-generation families, including newborns

Jung-Ha Kim

Department of Medicine
The Graduate School, Yonsei University

(Directed by Professor Duck-Chul Lee)

Aim: Leukocyte telomere length (LTL), as an indicator of biological aging, is influenced by both genetic and environmental factors. A recent study has reported stronger maternal than paternal inheritance. However, the extent of LTL heritability and inheritance patterns is still controversial. Newborns have not been included in any prior studies, and most studies have used Caucasian subjects. The aim of this study was to determine the heritability and inheritance patterns of telomere length across three generations of Korean families.

Methods: This cross-sectional study comprised 287 individuals from three generations of 41 Korean families, including newborns, parents, and maternal and paternal grandparents. LTL was measured using the telomere repeat copy number to single gene copy number (T/S) ratio as determined by quantitative real-time polymerase chain reaction. We estimate the heritability and inheritance patterns of telomere length among study participants after adjusting for potential confounders as well as for age and sex. Heritability was estimated by means of SOLAR software using the maximum-likelihood variance

component methods and a pedigree dataset. To evaluate the pairwise relationships between telomere lengths, Pearson's partial correlation with age adjustment was performed. The "cocor" package was used to compare LTL correlations based on dependent groups with overlapping variables. To evaluate whether the length of marriage was associated with the telomere length correlation between husbands and wives, Pearson's partial correlations with adjusted for age and length of marriage were performed in spousal pairs. We calculated Spearman's rank-order correlation coefficients between spousal rank differences of telomere length and length of marriage.

Results: Heritability of LTL in all participants was strong (h^2=0.64). There were no statistically significant differences in correlation coefficients of telomere length between paternal and maternal lines. There was a positive LTL correlation in grandfather-grandmother pairs (r=0.25, p=0.03), but not in father-mother pairs. After adjusting for age and length of marriage, the relationship between telomere lengths in grandfathers and grandmothers disappeared. There were inverse correlations between spousal rank differences of telomere length and length of marriage (all pairs ρ=-0.50, p <0.01; grandparents pairs ρ=-0.66, p <0.01; parents pairs ρ=-0.39, p <0.01).

Conclusions: These results show that LTL is both highly heritable without a sex-specific inheritance pattern and also influenced by shared environment.

Key words : Asian continental ancestry group, genetics, newborn, pedigree, telomere length.

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I. INTRODUCTION

Telomeres are repetitive sequences of DNA (TTAGGG), located at the ends of mammalian chromosomes, that play a role in maintaining genomic stability and function.[1] It is known that telomere length is influenced not only by birth telomere length as a genetic factor[2] but also by environmental factors such as smoking, sedentary lifestyle, and psychosocial status.[3] The establishment of the heritability and inheritance patterns of telomere length would represent an important cornerstone in understanding telomere length dynamics. In previous studies, the estimated heritability of telomere length ranged from 34% to 82% in humans.[4-8] One study showed that the rank of an individual's telomere length did not change over time.[9] It is still questionable to what degree genetic factors influence the telomere length of offspring. Inheritance patterns of telomere length also remain controversial. In an earlier study, it was reported that telomere length inheritance was linked to the X chromosome.[10] In addition, several studies have shown paternal inheritance.[11-13] On the other hand, stronger maternal inheritance rather than paternal inheritance was suggested in a recent

meta-analysis.[5] Another meta-analysis concluded that there is no significant difference between father-offspring and mother-offspring telomere length correlations.[14] Most of the aforementioned studies utilized Caucasian subjects; only a few studies have been conducted in Asia,[15,16] and no study has explored heritability in telomere length between parents and newborns. The purpose of the study was therefore to determine the heritability and inheritance patterns of telomere length by examining three generations of Korean families including newborns, parents, and maternal and paternal grandparents. Furthermore, the correlation between spousal pairs' telomere lengths was also evaluated in order to assess environmental influence on telomere length.

II. MATERIALS AND METHODS

1. Study participants

This family-based study included 287 individuals from 41 families, consisting of seven members per family (26 male/15 female newborns, parents, and paternal/maternal grandparents)(Figure 1).

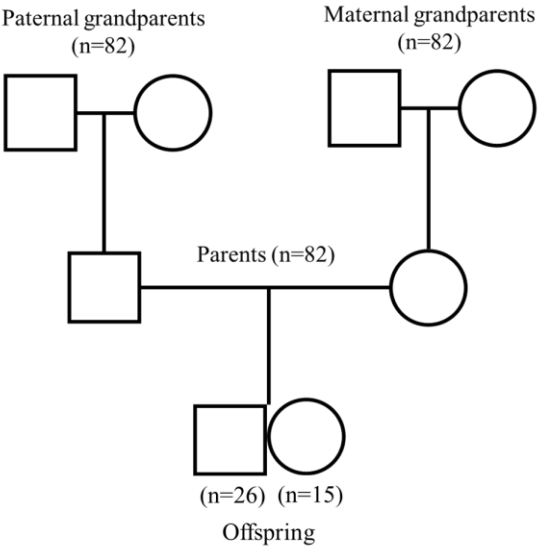

Figure 1. Study participants (n=287)

Healthy pregnant mothers in the 3rd trimester were recruited through an obstetrics and gynecology hospital and a university based childbirth clinic in Seoul between October 2012 and May 2013. A questionnaire survey, dietary interview, physical examination, and blood collection were performed. Pregnant women and husbands were excluded when they were self-reported to have been diagnosed by a medical doctor with hypertension, diabetes, dyslipidemia, cancer, chronic kidney disease, thyroid disease, cardiovascular disease (such as stroke or myocardial infarction), and liver disease (including B/C-viral hepatitis and liver cirrhosis). By the same token, grandparents were excluded when they were self-reported to have been diagnosed by a medical doctor with cancer, chronic kidney disease, thyroid disease, cardiovascular disease (such as stroke or

myocardial infarction), or liver disease (including B/C-viral hepatitis and liver cirrhosis). Participants were also excluded if pre-eclampsia, eclampsia, or gestational diabetes developed in the pregnant mother, if a congenital anomaly such as Down syndrome was suspected in the fetus by the triple test or fetal ultrasound, or if the newborn was born prematurely (prior to 37 weeks of pregnancy). At delivery, newborn cord blood was collected. The newborns' fathers and paternal and maternal grandparents were then asked to visit a family medicine clinic at Chung-Ang University Hospital for a medical examination between October 2013 and May 2014.

2. Questionnaires

Length of marriage, alcohol consumption, smoking status, physical exercise, current medications, and medical history were assessed using a questionnaire. Heavy alcohol consumption was defined as 15 drinks or more per week for men and 8 drinks or more per week for women. Current smokers were defined as those who were smoking at the time of the survey and had smoked more than five packs of cigarettes during their lifetime. Regular exercise was defined as physical exercise performed for at least 30 minutes more than three times per week. Quality of life was measured using the Korean version of the 36-item Short-Form health survey (SF-36).[17] Physical and mental component summary scores (from 1 [worst] to 100 [best]) were calculated based on the eight dimensions of the SF-36. Nutritional assessment was performed using dietary

intake data obtained using the 24-hour recall method. Dietary data was analyzed using CAN-pro 3.0 (Korean Nutrition Society, Korea), a professional software program used for nutrient calculation. Protein, fat, and carbohydrate intakes were presented as percentages of total energy consumption.

3. Physical examination and biochemical assay

Body weight (other than self-reported pre-pregnancy weights) was measured while participants wore light clothing and no shoes. Height was measured using a stadiometer. Body mass index (BMI) was calculated as weight divided by height squared (kg/m^2). Grip strength was measured using a strain-gauged dynamometer (Takeii TKK 501; Scientific Instruments Co. Ltd., Tokyo, Japan). Participants performed three trials on each hand, alternating sides. Maximal grip score was used to measure grip strength.[18] Biochemical tests were only performed on parents' and grandparents' blood samples collected after overnight fasting (> 12 hours). Fasting glucose, triglycerides, total cholesterol, and high-density lipoprotein (HDL) cholesterol levels were measured using enzymatic methods on an AU 5800 chemistry analyzer (Beckman Coulter, Brea, CA, USA). Low-density lipoprotein (LDL) cholesterol was calculated using Friedewald's formula (total cholesterol - HDL cholesterol - triglycerides/5). Total white blood cell (WBC) count was measured by flow cytometry using fluorescent dyes (XN-9000, SYSMEX, Kobe, Japan).

4. Measurement of leukocyte telomere length

Measurement of telomere length in leukocyte genomic DNA was accomplished by extraction of DNA from whole blood using the G-spin Genomic DNA Extraction Kit for Blood (iNtRON Biotechnology Inc, Gyeonggi-Do, Korea). All DNA samples were diluted to the same concentration (based on UV absorbance) and stored at -80℃ until time of use. As previously described, Leukocyte telomere length was measured as telomere repeat copy number to single gene copy number (T/S) ratio by quantitative real time polymerase chain reaction (PCR).[19] Real-time PCR was performed using a Light-Cycler 2.0 (Roche Diagnostics, Mannheim, Germany), and the rate of accumulation of amplified DNA was measured by continuous monitoring with the LightCycler® FastStart DNA Master SYBR Green I kit (Roche Diagnostics), with $MgCl_2$ at a final concentration of 2 mM. The primers for the telomere PCR were 200 nmol/L of 5'-GGTTTTTGAGGGTGAGGGTGAGGGTGAGGGTG AGGGT-3' and 200 nmol/L of 5'-TCCCGACTATCCC TATCCCTATCCCTA TCCCTATCCCTA-3'. The primers for the β-globin PCR were 300 nmol/L of 5'-GCTTCTGACACAACTGTGTTCACTAGC-3' and 500 nmol/L of 5'-CACC AACTTCATCCACGTTCACC-3'. The thermal cycling profile for telomere amplification was 10 minutes at 95℃ followed by 25 cycles at 95℃ for 10 seconds and one minute at 58℃. The thermal cycling profile for β-globin amplification was 95℃ for 10 minutes followed by 35 cycles at 95℃ for 10

seconds and at 56℃ for 15 seconds. Each sample was run in duplicate, using 25 ng of DNA per 10 μL reaction. A no-template control was included in each run, and the same calibrator sample was used in all runs to allow comparison of results across runs. Melting curve analysis was performed on every run to verify the specificity and identity of the PCR products. Quantitative values were obtained from the Ct values at which single increases associated with the exponential growth of PCR products were detected using LightCycler. The Ct values generated were used to calculate the T/S ratio for each sample using the following equation: $T/S = 2^{-\Delta Ct}$ (where $\Delta Ct = Ct_{single\text{-}copy\ gene} - Ct_{telomere}$). The coefficients of variation of the telomere, single gene, and T/S ratio duplicate assays were less than 4%, less than 3%, and less than 5%, respectively.

5. Statistical analyses

Age and leukocyte telomere length (T/S ratio) are presented as mean ± standard deviation (SD). Since the distribution of leukocyte telomere length was skewed, telomere length was logarithmically transformed. Telomere length according to generation and sex was compared by t-test or ANOVA.

Estimation of heritability (h^2) was performed using maximum-likelihood variance components methods implemented by the Sequential Oligogenic Linkage Analysis Routines (SOLAR) software (SOLAR Eclipse version 8.1.1; http://solar-eclipse-genetics.org/) using a pedigree dataset.[20] The variance components model assumes that the phenotypic variance of a given trait can be

partitioned into genetic and environmental components. Heritability was defined as the proportion of genetic variance to total phenotypic variance[21] using a maximum likelihood method applied to a mixed-effects model that incorporates fixed effects for known covariates and variance components for genetic effects. The significance of each heritability estimate was assessed using a comparison of the polygenic model to a sporadic model in which the additive genetic effect was constrained to zero.

Heritability (h^2, 95% confidence interval [CI]) was calculated with adjustment for age, sex, age×sex, age^2 and age^2×sex, or with adjustment for significant covariates in the best model after removal of covariates without significance ($p \geq 0.1$). Covariates included in the screening model were age; sex; BMI; heavy alcohol consumption; smoking; regular exercise; medical history of hypertension, diabetes, or dyslipidemia; dietary intake, including total energy, protein, and fat intake; grip strength; and fasting glucose, triglyceride, HDL-cholesterol, and WBC counts. The association between telomere length and covariates is presented as the beta coefficient ± standard error (SE). In addition, to examine paternal or maternal heritability separately, we assumed that one parent and one grandparent were known. For example, only phenotypic information on fathers and paternal grandfathers was provided for paternal heritability.[7]

To evaluate the pairwise relationships between telomere lengths, Pearson's partial correlation coefficients were calculated with age adjustments. For

multiple tests between newborn-pairs (mother, father, maternal grandmother, maternal grandfather, paternal grandmother, or paternal grandfather), p-values were corrected using the Bonferroni method. The cut-off for the p-value was 0.05/6.

We evaluated whether the length of marriage in spousal pairs was associated with telomere length correlations between husbands and wives. Pearson's partial correlations with adjustments for age and length of marriage were performed in spousal pairs. We then ranked telomere length separately according to sex. We calculated Spearman's rank-order correlation coefficients between spousal rank differences in telomere length and length of marriage (adjusted for age).

Pearson's partial and Spearman's rank-order correlations were performed using the SAS 9.1 statistics package (SAS Institute, Inc., Cary, NC, US). The cocor package (http://comparingcorrelations.org/) was used for comparing two correlations based on dependent groups with overlapping variables.[22]

III. RESULTS

1. General characteristics of study participants

The mean ages of fathers, mothers, grandfathers, and grandmothers were 34.9 years, 32.8 years, 64.9 years, and 61.9 years, respectively. Tables 1 and 2 show the general characteristics of the participants. The average length of marriage was 3.1±1.4 years in parents and 33.1±4.9 years in grandparents.

Table 1. General characteristics of parents

	Father (n=41)	Mother (n=41)
Age (years)	34.9±3.5	32.8±2.9
Body weight (kg)	78.0±11.7	54.3±7.0
Height (cm)	174.4±5.1	160.4±4.9
Body mass index (kg/m^2)	25.5±4.1	21.0±2.5
Systolic blood pressure (mmHg)	129.0±15.7	111.1±12.3
Diastolic blood pressure (mmHg)	77.9±11.0	68.9±10.4
Grip strength, left (kg)	39.8±9.4	22.8±4.5
Grip strength, right (kg)	40.7±9.8	23.4±5.0
Fasting glucose (mg/dL)	98.4±10.5	97.9±22.9
Total cholesterol (mg/dL)	195.9±40.7	246.7±56.9
Triglycerides (mg/dL)	178.8±90.6	247.4±108.5
High density lipoprotein-cholesterol (mg/dL)	48.6±9.2	66.9±15.2
White blood cells (/μL)	5976.1±1360.2	10389.1±4153.2
Nutrient intake		
Total energy (kcal/d)	2054.6±224.7	1656.8±358.8
Protein (% of energy intake)	16.5±2.4	16.9±5.4
Fat (% of energy intake)	27.4±4.9	19.4±5.0
Carbohydrate (% of energy intake)	57.0±6.4	65.0±6.4
QoL: SF-36 mental component score	70.9±10.4	73.8±13.5
QoL: SF-36 physical component score	79.9±11.7	70.2±9.8
Current smoker, n(%)	6(14.6)	2(4.9)*
Alcohol drinking, n(%)	30(73.2)	15(36.6)*
Regular exercise, n(%)	15(36.6)	12(29.3)

QoL, quality of life

*Before pregnancy

Table 2. General characteristics of grandparents

	Grandfather (n=82)	Grandmother (n=82)
Age (years)	64.9±4.9	61.9±4.7
Body weight (kg)	68.9±9.6	60.6±9.0
Height (cm)	166.0±5.9	154.1±4.4
Body mass index (kg/m^2)	25.1±3.2	25.6±3.5
Systolic blood pressure (mmHg)	136.7±12.8	129.8±16.9
Diastolic blood pressure (mmHg)	80.8±8.3	76.5±9.5
Grip strength, left (kg)	34.3±8.0	21.8±4.9
Grip strength, right (kg)	35.1±7.8	22.9±4.8
Fasting glucose (mg/dL)	121.3±37.8	104.6±23.0
Total cholesterol (mg/dL)	190.0±37.5	194.4±40.4
Triglycerides (mg/dL)	167.8±92.6	108.1±52.3
High density lipoprotein-cholesterol (mg/dL)	48.5±10.0	52.2±8.5
White blood cells (/μL)	5869.4±1876.3	5447.7±1355.5
Nutrient intakes		
Total energy (kcal/d)	1808.6±239.3	1499.3±203.5
Protein (% of energy intake)	16.8±3.0	15.9±2.2
Fat (% of energy intake)	20.8±3.7	19.3±5.6
Carbohydrate (% of energy intake)	62.0±4.8	65.3±6.5
QoL: SF-36 mental component score	51.8±7.2	47.8±6.9
QoL: SF-36 physical component score	42.6±8.7	42.8±7.8
Current smoker, n(%)	23(28.0)	2(2.4)
Alcohol drinking, n(%)	28(34.1)	5(6.1)
Regular exercise, n(%)	30(36.6)	36(43.9)
Hypertension*	24(29.3)	25(30.5)
Diabetes†	16(19.5)	8(9.8)
Dyslipidemia‡	7(8.5)	10(12.2)

QoL, quality of life

*Self-reported hypertension, or ≥140 mm Hg for the systolic measurement or ≥90 mmHg for the diastolic measurement

†Self-reported diabetes or fasting glucose ≥126 mg/dL

‡Self-reported dyslipidemia, total cholesterol ≥240 mg/dL, LDL-cholesterol ≥160 mg/dL, or triglycerides ≥200 mg/dL

2. Mean telomere length of all participants

There were significant differences the mean leukocyte telomere length among the three groups of participants (newborns: 1.65 ± 0.65, parents: 1.18 ± 0.48, and grandparents: 0.87 ± 0.32, $p < 0.01$). Telomere length was greater in mothers (1.34 ± 0.51) and grandmothers (0.95 ± 0.34) than in fathers (1.01 ± 0.40) and grandfathers (0.79 ± 0.29), respectively (all $p < 0.01$, Figure 2).

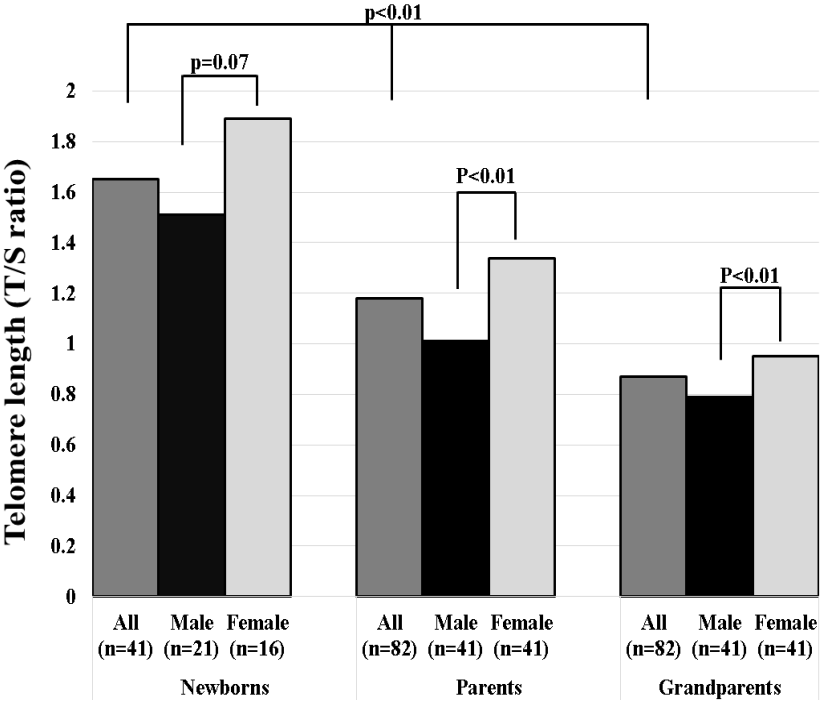

Figure 2. Telomere length according to generation and sex of study participants. P-values were calculated by t-test or ANOVA. T/S ratio: ratio of telomere repeat copy number to single gene copy number.

3. Estimated heritability (h^2) of telomere length

The covariates of age×sex (p=0.34), age^2 (p=0.33), and age^2×sex (p=0.24) were removed from the final analytic model. Leukocyte telomere length was significantly associated with age (β=-0.022 ± 0.001) and sex (β=0.373 ± 0.088) (all $p < 0.01$). Age and sex-adjusted heritability of telomere length in all family relations was 0.64 (95% CI 0.47-0.81, $p < 0.01$). Heritability estimates according to generation and sex are shown in Table 3. There was no difference in the heritability estimates between male (0.58, 95% CI 0.31-0.86, $p < 0.01$) and female newborns (0.62, 95% CI 0.35-0.89, $p < 0.01$)(t=0.17, p=0.86).

When we adjusted for potential confounders, the significant variables included in the best model were age (β=-0.030 ± 0.003, $p < 0.01$), sex (β=1.007 ± 0.156, $p < 0.01$), grip strength (β=0.059 ± 0.008, $p < 0.01$), and dietary fat intake (β=-0.011 ± 0.003, p=0.01) in the grandparents-parent trios. In the grandmother-parent pairs, age (β=-0.027 ± 0.003, $p < 0.01$), grip strength (β=0.007 ± 0.006, $p < 0.01$), and SF-36 mental component summary score (β=0.018 ± 0.006, p = 0.04) were included in the best model. In the grandfather-parent pairs, age (β=-0.010±0.008, p = 0.02), sex (β=1.290 ± 0.457, $p < 0.01$), BMI (β=-0.029 ± 0.019, p = 0.02), grip strength (β=0.072 ± 0.013, $p < 0.01$), SF-36 mental component summary score (β=0.005 ± 0.007, p = 0.06), dietary fat intake (β=-0.008 ± 0.007, p = 0.02), current smoking (β=-0.329 ± 0.263, p = 0.09), and hypertension (β=-0.211 ± 0.139, p = 0.01) were also included.

Table 3. Estimated heritability (h^2) of telomere length

	Age and sex -adjusted		Covariates -adjusted	
	h^2 (95% CI)	P-value	h^2 (95% CI)	P-value
Grandparents-parents-newborn family (n*=41)	0.64 (0.47-0.81)	0.001		
Paternal grandfather-father -newborn trio (n=41)	0.80 (0.37-1.23)	0.001		
Maternal grandmother-mother -newborn trio (n=41)	0.85 (0.52-1.18)	0.001		
Grandparents-parent trio (n=82)	0.56 (0.38-0.74)	0.001	0.55 (0.38-0.72)†	<0.001
Grandfather-parent pair (n=82)	0.58 (0.36-0.80)	0.004	0.56 (0.36-0.76)‡	<0.001
Grandmother-parent pair (n=82)	0.69 (0.52-0.86)	0.001	0.60 (0.42-0.78)§	0.004
Parents-newborn trio (n=41)	0.68 (0.53-0.84)	<0.001	-	-
Father-newborn pair (n=41)	0.76 (0.60-0.92)	0.001	-	-
Mother-newborn pair (n=41)	0.89 (0.75-1.03)	<0.001	-	-

*Number of pairs, trios, or families

†Age, sex, grip strength, and dietary fat intake were included as the covariates in the analytic model

‡Age, sex, body mass index, grip strength, SF-36 mental component summary score, dietary fat intake, current smoking, and hypertension were included as covariates in the analytic model

§Age, grip strength, and SF-36 mental component summary score were included as covariates in the analytic model

4. Pairwise relationships between telomere lengths

A. The relationship between telomere lengths of newborns and parents

Telomere lengths of newborns were only positively associated with those of their mothers (r=0.63, p < 0.0001)(Figure 3).

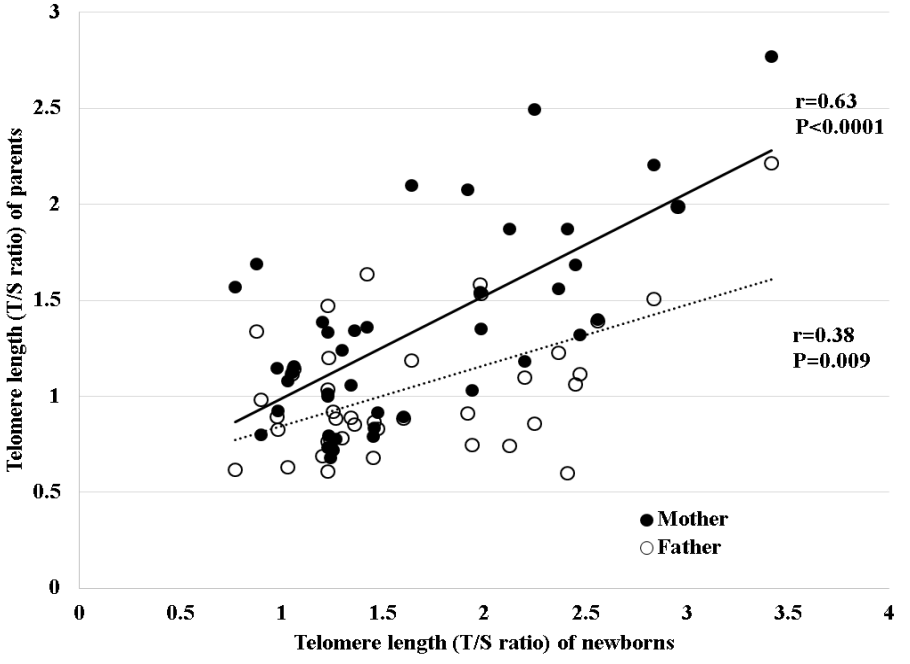

Figure 3. Correlation of telomere lengths between newborns and parents. Pearson's partial correlation coefficients were calculated with age adjustments. For multiple tests, significant level was corrected using the Bonferroni method 0.05/6.

B. The relationship between telomere lengths of newborns and grandparents

Telomere lengths of newborns were not correlated with those of paternal (Figure 4) and maternal (Figure 5) grandparents.

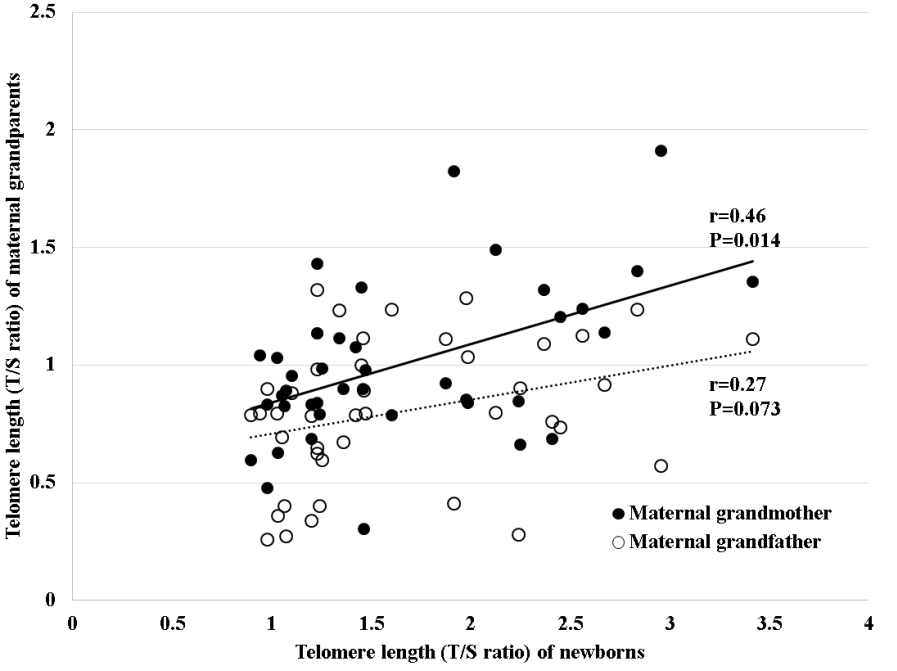

Figure 4. Correlation of telomere lengths between newborns and maternal grandparents. Pearson's partial correlation coefficients were calculated with age adjustments. For multiple tests, significant level was corrected using the Bonferroni method 0.05/6.

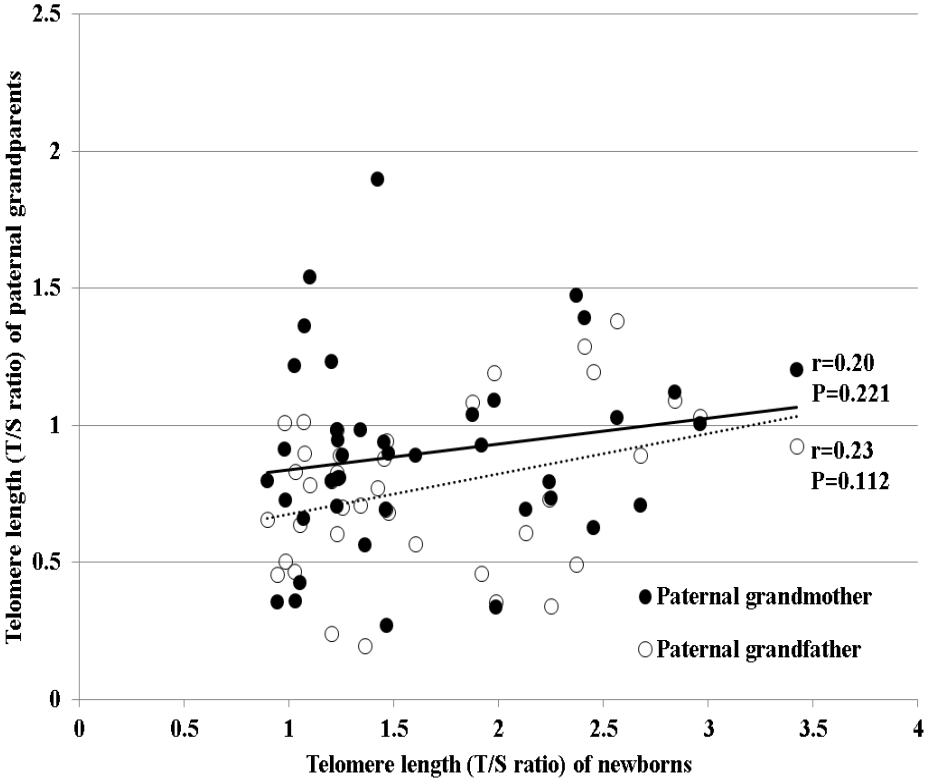

Figure 5. Correlation of telomere lengths between newborns and paternal grandparents. Pearson's partial correlation coefficients were calculated with age adjustments. For multiple tests, significant level was corrected using the Bonferroni method 0.05/6.

C. The relationship between telomere lengths of parents and grandparents

There was a positive association between the telomere lengths of parents and grandparents (grandmothers: r=0.35, p < 0.01; grandfathers r=0.23, p=0.04) (Figure 6). However, there were no significant differences in the correlation coefficients of telomere length between paternal and maternal lines (Table 4).

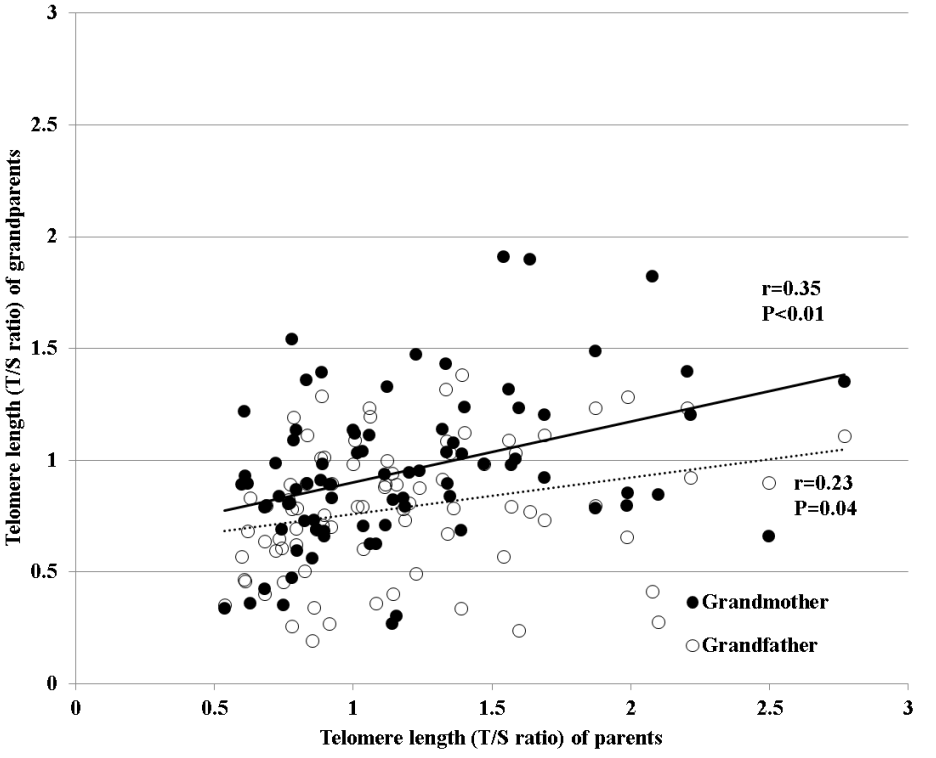

Figure 6. Correlation of telomere lengths between parents and grandparents. Pearson's partial correlation coefficients were calculated with age adjustments.

Table 4. Differences in correlation coefficients of telomere length between paternal and maternal pairs

	Z^{*}	$t^{\dagger}$	Confidence interval[‡]	*P*-value[§]
Grandfather-parent vs. grandmother-parent	0.90	0.94	0.13~0.37	0.35
Father-newborn vs. mother-newborn	1.55	1.64	0.07~0.58	0.12
Paternal grandfather-newborn vs. paternal grandmother-newborn	0.16	0.16	0.34~0.40	0.88
Maternal grandfather-newborn vs. maternal grandmother-newborn	1.11	1.10	0.16~0.53	0.27
Paternal grandfather-newborn vs. maternal grandfather-newborn	0.20	0.20	0.36~0.44	0.85
Paternal grandmother–newborn vs. maternal grandmother-newborn	1.38	1.37	0.13~0.63	0.17

[*]by Pearson and Filon's method.

[†]by Hotelling's method.

[‡]by Zou's method.

[§]A comparison (two-sided, alpha = 0.05, confidence level = 0.95) using the cocor package of two overlapping correlations based on dependent groups.

D. The relationship between telomere lengths of husbands and wives

(A) Without consideration of length of marriage

After age adjustment, there was only a positive association between the telomere lengths of grandfathers and grandmothers (r=0.25, p=0.03)(Figure 7).

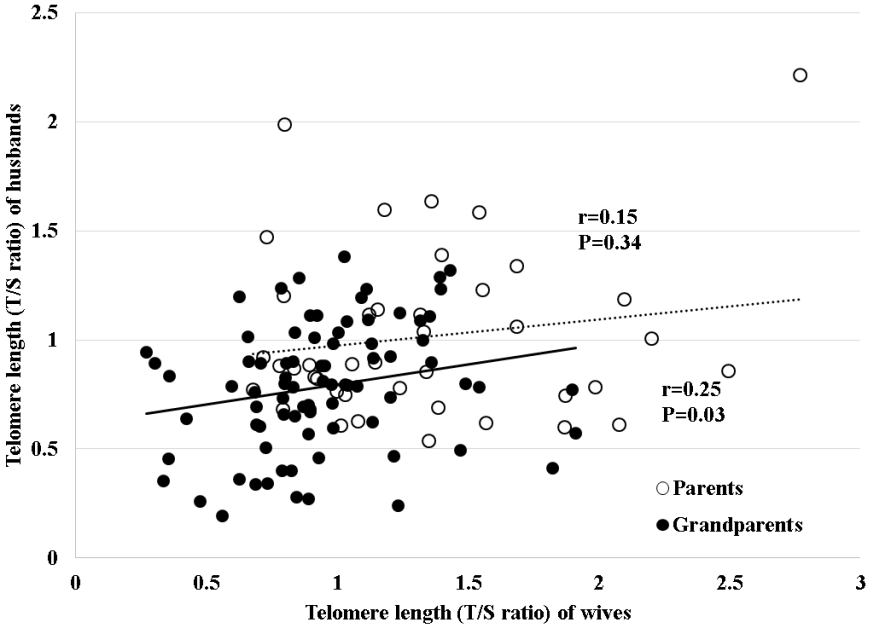

Figure 7. Correlation of telomere lengths of husbands and wives. Pearson's partial correlation coefficients were calculated with age adjustments.

(B) With consideration of length of marriage

After adjusting for age and length of marriage, the relationship between the telomere lengths of grandfathers and grandmothers disappeared (r=0.20, p=0.07). After age adjustment, there were inverse correlations between spousal

rank differences of telomere length and length of marriage (all pairs ρ=-0.50, p <0.01; grandparents pairs ρ=-0.66, p <0.01; parents pairs ρ=-0.39, p <0.01).

IV. DISCUSSION

This three-generation (including newborns) family-based study demonstrated a high estimated telomere length heritability of 0.64, without a sex-specific inheritance pattern. There was a positive correlation between the telomere lengths of husbands and wives in the older generation, but not in the younger generation. The length of marriage has been shown to influence the relationship between the telomere lengths of husbands and wives.

Several studies have shown a wide range of estimated telomere length heritabilities and inconsistent inheritance patterns. In one study, paternal inheritance of telomere length was reported among independent pairs or trios selected from 2~5-generation families (ages 12~102 years).[11] Another study of 132 healthy individuals, including off-spring (aged 32 to 42 years) and their parents (aged 52 to 86 years), suggested paternal inheritance of telomere length.[12] A third study also showed paternal inheritance among individuals with low bone mineral density or history of fracture, as well as in their spouses and all first-degree relatives (18~92 years; mean age of 49±17 years).[13] The latter study reported a relatively low estimated heritability of 0.44. The others did not report an estimated heritability. On the other hand, a meta-analysis of six large, independent cohort studies with a total of 19,173 participants aged 15-99 years

showed a high heritability of 0.70, with a maternal inheritance pattern.[5] Possible explanations for these dominant sex-specific parental effects on telomere length inheritance have been suggested.[23,24]

However, recently it has been demonstrated that, as shown in our results, the heritability of telomere length dose not differ between paternal and maternal lines. A meta-analysis within a population of European descendants showed no significant differences in telomere length correlations between mother-offspring and father-offspring.[14] A large study of long-lived families showed an overall heritability of 0.54 between long-lived elderly and their offspring (mean age of 61.7 years), adjusting for multiple demographic and environmental factors. Heritability of telomere length did not differ between male parents and their offspring and female parents and their offspring.[7] These results suggest that the telomere length of newborns may be determined by genetic factors rather than by epigenetic or non-genetic (maternal environment) factors.

There is also a race/ethnic difference in telomere length.[25] In a study of Arabs, heritability between individual parents and their offspring (n=42) was estimated at 0.64, as in our results, but offspring telomere length was more highly correlated with paternal telomere length than with maternal telomere length.[15] Longer maternal telomere length predicted longer offspring telomere length in the Philippines.[16] However, heritability estimates in these studies do not involve additive genetic and parental effects, since they were suggested through a parent-offspring regression model. The latter study[16] did not examine an

association between paternal telomere length and offspring telomere length.

Two previous studies have shown conflicting results with regard spousal correlations in telomere length. One study reported no significant association between spouses aged 52-86 years.[12] Another study reported a significant association between spouses that was stronger in older couples (mean age ≥55years) than in younger couples (mean age <55years).[5] We also found a significant telomere length relationship between husbands and wives in their sixties. Telomere length between young couples in their early thirties was not correlated. Telomere length may also reflect environmental effects.[3]

The current study has a few notable strengths. As far as we know, this is the first study identifying heritability and inheritance patterns of telomere length in parents-newborn trios. We included newborns in order to minimize environmental influences in order to arrive at a more precise estimation of heritability and a more accurate evaluation of inheritance patterns in telomere length. Only one British study including telomere length measured at childbirth showed a highly significant correlation between maternal and newborn telomere length, without reports of heritability of telomere length or a correlation between paternal and newborn telomere length.[26] In addition, our study estimated the heritability of telomere length in East Asians, who differ genetically, culturally, and environmentally from previous Caucasian study populations, using extended pedigrees. The present study also evaluated various confounding factors affecting telomere attrition, examining anthropometric

measurements, grip strength, inflammatory markers, psychological factor, health behaviors, medical history, and nutritional intake.

Several limitations should also be noted. First, this study is not a population-based study but instead is small, selective, and ethnically homogenous, which limits the generalizability of its results. Second, the small sample size may make it more difficult to discern differences between heritability estimates by sex or generation. Third, we could not adjust telomere length according to WBC count because analysis of newborns' WBC counts was not performed. Recent studies have shown that telomere length is positively associated with WBC counts.[27] Another issue was that blood from parents was collected at different times. Mothers' blood samples were collected during pregnancy, while fathers' blood samples were collected after their children were born. These factors could influence the mother-newborn relationship in telomere length. Finally, we did not examine certain demographic factors such as educational level, residential area, and occupation. Recent studies have reported that maternal education level[28] and residential traffic exposure[29] are associated with telomere length at birth. These demographic factors could therefore be significant environmental components of the estimated heritability of telomere length.

V. CONCLUSION

In conclusion, it appears that telomere length has high heritability and does not show a sex-specific inheritance pattern. Given the differences in spousal telomere length relationships between generations, it seems that telomere length might also be influenced by environmental factors. Further studies in East Asian populations are needed to clarify these heritability and inheritance patterns of telomere length and to further identify the factors that influence them.

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ABSTRACT(IN KOREAN)

한국 3세대 직계 가족에서의 텔로미어 길이 유전력

<지도교수 이덕철>

연세대학교 대학원 의학과

김정하

목적: 텔로미어 길이는 세포 노화의 지표로 유전과 환경에 영향을 받는 것으로 알려져 있다. 그러나, 텔로미어 길이의 유전력과 그 패턴에 대해 아직 논란이 있으며, 주로 서양인을 대상으로 연구되어 동양인에서는 이에 대한 연구가 드물다. 이 연구에서는 신생아를 포함한 한국인 3대 가족을 통해 백혈구 텔로미어 길이의 유전력과 유전 패턴에 대해 알아보고자 하였다.

방법: 신생아와 부모, 친·외조부모 7명으로 구성된 41가족 287명을 대상으로 하였다. 백혈구 텔로미어 길이는 실시간 PCR로 베타-글로빈 유전자 카피 수에 대한 텔로미어 카피 수의 비로 측정하였다. 유전력은 SOLAR 소프트웨어를 이용하여 추정하였다. 부계와 모계 각각에 따른 텔로미어 길이 상관계수는 나이를 보정한 편상관관계분석을 통해 구하였고, 상관계수의 비교는 cocor 패키지를 이용하였다. 결혼기간이 부부간 텔로미어 길이 연관성과 관련되는지 확인하기 위해 나이와 결혼기간을 보정한 편상관관계분석을 시행하였다. 또한 성별에 따라 텔로미어 길이 순위를 매겨 부부간 텔로미어 길이 순위 차이와 결혼기간과의 상관성을 알아보기 위해 스피어만 서열상관계수를 구하였다.

결과: 백혈구 텔로미어 길이의 유전력은 0.64였다. 부계와 모계 사이의 상관계수의 차이는 없었다. 조부모 사이에 텔로미어 길이는 양의 상관관계를 보였으나($r=0.25$, $p=0.03$), 이들에서 결혼기간 보정 후에는 유의한 상관관계를 보이지 않았다($r=0.20$, $p=0.07$). 부부간 텔로미어 길이의 순위 차이와 결혼기간은 음의 상관관계를 보였다(모든 부부: $\rho=-0.50$, $p<0.01$; 조부모: $\rho=-0.66$, $p<0.01$; 부모 $\rho=-0.39$, $p<0.01$).

결론: 백혈구 텔로미어 길이는 강한 유전력을 보였으며, 부계나 모계의 성별에 특정된 유전 패턴을 나타내지는 않았다. 또한 부부간 텔로미어 길이의 상관성이 결혼기간과 관련되는 것으로 보아 환경의 영향을 받는 것으로 여겨진다.

핵심되는 말 : 가계, 동양인, 신생아, 유전, 텔로미어 길이

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