ASIAN JOURNAL OF PHARMACEUTICAL AND CLINICAL RESEARCH

INNOVARE ACADEMIC SCIENCES
Knowledge to Innovation

Vol 11, Issue 7, 2018

Online - 2455-3891
Print - 0974-2441

Research Article

THE CORRELATION BETWEEN ESTRADIOL LEVELS WITH TELOMERE LENGTH BASED ON HISTORY OF USING HORMONAL CONTRACEPTIVE IN PREMENOPAUSAL WOMEN

YUSRAWATI[1], DELMI SULASTRI[2], DESMAWATI[2]

[1]Department of Obstetrics and Gynecology, Faculty of Medical, University of Andalas, Kota Padang 25163, Sumatera Barat, Indonesia.
[2]Department of Nutrition, Faculty of Medical, University of Andalas, Kota Padang 25163, Sumatera Barat, Indonesia.
Email: yusrawati65@med.unand.ac.id/ desmawati@med.unand.ac.id

Received: 07 March 2018, Revised and Accepted: 14 April 2018

ABSTRACT

Objective: This study aimed to investigate the correlation between nutritional status and estradiol level with telomere length in premenopausal women with and without hormonal contraceptive history.

Methods: This cross-sectional study was conducted in Padang city to 115 premenopausal Minangkabau ethnic 40–54 years old. Height examined using stature meter, waist circumference (WC) using measuring tape, and body weight using body scale were performed by trained people. Blood sample analysis was performed using O'Callaghan and Fenech's technique to measure telomere length. Estradiol level was measured by enzyme-linked immunosorbent assay technique.

Results: This research indicated that an average telomere length was 552.96±422.57 bp, body mass index (BMI) was 26.12±4.84, WC was 85.66±10.28 cm, and estradiol level was 148.24±54.03 pg/ml. The proportion of subjects who have hormonal contraceptives history was 65.2% with the most method of contraceptive used injection (75%). There were no correlations between BMI, WC, and estradiol level with telomere length ($p>0.005$). However, there was a positive significant correlation between estradiol levels and telomere length in women with no history of hormonal contraceptive use ($p=0.025$).

Conclusion: A history of hormonal contraceptive used influences the correlation between estradiol levels and telomere length in premenopausal women.

Keywords: Telomere length, Body mass index, Estradiol, Hormonal contraceptive, Telomere shortening.

 DOI: http://dx.doi.org/10.22159/ajpcr.2018.v11i7.25671

INTRODUCTION

Hormonal contraceptives are used widely to prevent the pregnancy in productive age. Approximately 15–20% of contraceptive users used hormonal contraceptives, especially in developing countries. In Indonesia, in 2015, the hormonal contraceptives used were 49% of total contraceptive users, and injections are the widest method [1]. Hormonal contraceptives generally contain estrogen, progesterone, or a combination of both that affects ovulation, hormonal balance, and menstrual cycle [2]. However, the hormonal contraceptives used can cause some effects on both nutritional status and blood pressure. Later, it is known that estrogen has a role in inhibiting telomere restriction process. The telomere is an aging biomarker, in the sequence of nucleotide bases repeatedly located on the end of the chromosome. Estrogen is a female hormone that has antioxidant properties that can suppress the production of reactive oxygen species (ROS) and function as an antioxidant gene regulator [3]. Estrogen directly activates telomerase promoters and indirectly also affects the improvement of deoxynucleotide acid (DNA) through the P53 pathway and promoter activation through phosphoinositol-3-kinase/Akt and nitric oxide pathways. This leads to higher telomerase activity in women. [3]. The use of long-term hormonal contraceptives affects estrogen 2 (estradiol) levels in the body. Hence, we hypothesize that the use of hormonal contraceptives affects the telomere length.

In premenopausal, there is a decrease in estrogen production, especially estrogen 2 (estradiol). The estradiol levels in premenopausal are 15–350 pg/mL, whereas in menopausal women, it is lower than 10 pg/mL [4]. Along with this decline in estrogen hormone levels, the telomeres shortening process is accelerated, leading to increased degenerative disease in premenopausal women [5]. The effect of estrogen on telomere length has been demonstrated by studies, suggesting that postmenopausal women who use hormone replacement therapy have longer telomeres than those not treated [6].

Telomere length in humans varies between 5 and 15 kb, longer in females than males. The female telomere length is 7.01 kb while the male telomere length is 6.87 kb. Research in China suggests that female telomeres are longer than men in Tibetan and Han ethnic, but there is no difference in telomeres' lengths for both ethnic groups [7]. A 12-year longitudinal study conducted on 405 women and 329 men concluded that the shortening of the telomere in women was hormonally affected, i.e., estrogen. Other studies suggest that nutritional status also affects the telomere length. Obese person has shorter telomere than normal weight [8]. This study aims to investigate the correlation between nutritional status and estradiol level with telomere length in premenopausal women with and without hormonal contraceptive history.

METHODS

Selection of sample

This study was a cross-sectional study in 115 premenopausal women at Padang District, West Sumatera, Indonesia. All samples were Minangkabau ethnicity, 40–54 years old, selected randomly and signed an informed consent sheet. Women who have stopped menstruation for more than 12 months or use hormonal contraceptives in the past 5 years or have chronic illnesses such as diabetes mellitus, stroke, cardiovascular disease, and cancer are excluded from the study. Subjects with severe stress (selected by questionnaire) and athletes were also excluded.

Nutritional status examinations

Height examined using stature meter, waist circumference (WC) using a measuring tape, and body weight using body scale were performed

by trained people, done twice. Body mass index (BMI) was calculated based on weight in kg and height in m.

Blood collection and laboratory examination

Telomere examination was done from 3 ml of venous blood. First, we performed DNA isolation and then telomere length examination by using of O'Callaghan and Fenech method modification [9]. Estradiol level was measured by enzyme-linked immunosorbent assay technique. This study has had Ethical Approval from Ethical Committee Medical Faculty of Andalas University with registration number 279/KEP/FK/2017.

Statistical analysis

Data were analyzed using nonparametric Spearman's Rank correlation test with data split by hormonal contraceptives used history. A significant correlation considered when p-values <0.05. All data were analyzed using the SPSS software (version 22.0 for Windows, IBM Corp., NY, Armonk).

RESULT

115 premenopausal women have the characteristic as presented in Table 1.

From Table 1, we can see that the mean of BMI, WC, and telomere length is higher in women who do not have a history of hormonal contraceptive use compared with those who have a history, while estradiol levels were lower in those who did not have a history of hormonal contraceptive use.

Tables 1 and 2 show that most subjects having obese and central obesity and have a history of hormonal contraceptive use. The most common type of hormonal contraceptive used is injection.

The result of statistical analysis showed that there was no significant correlation between nutritional status, WC, and estradiol level with telomere length of research subjects. However, if data were analyzed based on a history of hormonal contraceptive use, then there was a positive significant correlation between estradiol levels and telomere length in women with no history of hormonal contraceptive use.

DISCUSSION

From Table 1, we can see that the mean of BMI, WC, and telomere length is higher in women who do not have a history of hormonal contraceptive use compared with those who have a history, while estradiol levels were lower in those who did not have a history of hormonal contraceptive use. This result is similar to Changamma's study that says that exogens hormonal administrations be able to increase the estradiol levels. In that study was explained that higher estradiol levels prevent gonadotropin-releasing hormone (GnRH) production in the hypothalamus, leading to a decrease in Follicle-stimulating hormone (FSH) production [10]. This is in contrast to Fleishchman's statement, in his article, stating that oral contraceptive suppresses ovarian hormone production including estradiol. however, in this study subjects were users of all hormonal contraceptives, not only oral contraceptives, even the most used injection contraception. This may be the cause of the difference in results [2]. Different ways of using hormonal contraceptives have different effects. Oral hormonal contraceptives may interact with food in absorption levels, whereas injections may be altered on its pharmacodynamic pathway [11].

The result showed that the mean of telomere length of these premenopausal women 552.96±422.57 bp is ranged from 79.33 bp to 2048.77 bp. This result is different in telomere length that found by Ochallagan and Fenech. They found the longer telomeres length with mean 941.30 bp (380.43–1891 bp) but it regardless of gender [9]. Other study in China got that female telomere length is 7.01 kb [7]. Many factors affect the telomere length. Age is an important factor because telomere shortening occurs every time cell division. In this study, subject has a wide range of age 40–54 years. This can cause a wide range of telomere lengths. Age has a negative correlation with telomere length, where 1 year of age increases, then telomere length decreases as much as 0.058 kb [12]. Laboratory methods also affect telomere length. In this study, the telomere length examination using the) quantitative polymerase chain reaction (qPCR) method resulted in the absolute telomere length (aTL), but most other studies used qPCR method which resulted in relative telomere length [13]. The telomere length relative of O'Challagan's research has an average of about 5000bp [9]. If this study's result were compared with the standard length of O'Callaghan, it is estimated that the mean of relative telomere length is about 3700bp. Sex also affects the telomere length. Gu *et al*. research found that female's telomere length was more than 129 bp than men [14]. Similarly, the Tiainen *et al*. study of 57– 70-year-old subjects obtained relatively long telomere length in women (1.41 kb) than in men (1.36 kb) [15]. Boccardi also found longer telomeres in females (5.06 kb) than males (4.73 kb) in aged 71–87 years [12]. Race also affects the telomere length. Telomere length examination on Hispanic race, African American race, and Caucasian gave different results. This study was conducted on Minangkabau ethnic so that telomere length cannot be compared with other research. In other words, telomere length on one ethnicity cannot be generalized to other ethnicities [15,16].

All subjects in this study were premenopausal women. At premenopausal, there is a decrease in estradiol production where estradiol levels are 15–350 pg/ml [4]. This study obtained that estradiol level of subject is 3.36–364.15 pg/ml with mean 148.24±54.03 pg/ml. Premenopausal women have greater BMI and WC also. Subject characteristic based on nutritional status, WC, and history of hormonal contraceptive use can be seen in Table 2.

Obesity and central obesity are risk factors for many diseases. Excessive intake of foods, especially high-fat diet with less physical activity, causes

Table 1: Average age, BMI, WC, estradiol levels, and telomere length of premenopausal women (n=115)

Variable	All			History of hormonal contraceptives used (-)			History of hormonal contraceptives used (+)		
	Mean±SD	Minimum value	Maximum value	Mean±SD	Minimum value	Maximum value	Mean±SD	Minimum value	Maximum value
Age (years)	46.80±3.87	40	54	47.2±3.91	41	54	46.6±3.85	40	54
BMI (kg/m^2)	26.12±4.84	16.17	43.76	26.78±5.88	16.44	43.2	25.77±4.18	16.17	40.04
WCs (cm)	85.66±10.28	56	119	85.92±11.67	66	119	85.52±9.52	56	108
Estradiol levels (pg)	148.24±54.03	3.36	364.15	139.20±48.23	63.14	230.87	153.06±56.59	3.36	364.15
Telomere Length (bp)	552.96±422.57	79.33	2048.77	594.34±416.77	103.18	1572.20	530.89±426.77	79.33	2048.77

BMI: Body mass index, WCs: Waist circumferences, SD: Standard deviation

fat accumulation, especially in the abdominal area. This fat accumulation will increase the release of ROS. Several studies say that BMI and WCs are associated with increased levels of ROS and DNA damage markers. Continuous oxidative stress will cause damage to cells, tissues, and organs that eventually lead to degenerative diseases [17,18].

In this study, it was found that 65.2% of subjects had a history of hormonal contraceptive use. In this study, 75% of subjects had a history of hormonal contraceptive use by injection. Hormonal contraceptives generally contain estrogen, progesterone, or a combination of both. Hormonal contraceptives have some negative effects, including increased weight, blood pressure, and associated with the decrease of renal function [19,20]. Others side effects are causing disruption to hair growth, allergies, affecting mood, and even some that cause thromboembolic disorder [21].

This study result suggests that there is no correlation between BMI and WC with telomere length in premenopausal (Table 3). This result is different from some previous studies. Ornish *et al*. suggested that telomere's erosion increases with weight gain in obese people. Similarly, Lee *et al*.'s study on non-Hispanic whites states that BMI has a negative association with telomere length. However, this result was similar with a recent study in Minangkabau man, and Sulastri *et al*. found that there is no association between BMI and telomere length [22].

Estradiol levels are also unrelated to telomere length. However, in Table 3, we can see that estradiol levels have a significant correlation with telomere length in women who have no history of hormonal contraceptive used. Long-term use of hormonal contraceptives affects endogenous estradiol levels and acts as LH receptor and progesterone hormone receptor inhibitors so that caused the premature ovulation or delayed ovulation [21]. Estradiol is an active estrogen produced by the ovaries during reproductive life. Estradiol has a protective effect against autoimmune diseases, Alzheimer's, cardiovascular disease, and osteoporosis. Estradiol also improves serotonin function so as to improve mood and improve cognitive function and emotional stability [23]. In premenopausal, there is a decrease in estradiol levels. This decrease in estradiol levels is associated with telomere length. Estrogen stimulates the activity of the enzyme telomerase directly by stimulating the activity of the TERT gene promoter. Indirectly, estrogen stimulates the expression of c-myc transcription factor. C-myc mediates the aurora-A kinase that induces telomerase activity [24]. Estrogen deficiency inhibits telomerase activity with decreased cell differentiation and atrophy of the adrenal gland [25]. *In vitro* studies suggest that increased telomerase activity and TERT gene expression are affected by estrogen binding with estrogen alpha receptors [26]. Hence, estrogen levels play a role against the telomere shortening rate. A decrease in estrogen levels in postmenopausal causes shorter telomeres shortening [5].

There are several limitations of this study, including cross-sectional design that cannot determine the causality relationship between variables. Then, the researchers did not consider other factors such as lifestyle and did not consider endogenous antioxidant level which also had an effect on maintaining the telomere. This study suggests that the person who has a history of hormonal contraceptive use should maintain a diet and lifestyle that affects the telomere level estradiol and length. To the best of our knowledge, this is the first study to discuss the history of hormonal contraceptive use that affects estradiol levels of telomere length relationship.

Table 2: Distribution of subjects based on nutritional status, WC, and history of hormonal contraceptive use (n=115)

Variables	n (%)
Nutritional status	
Underweight	5 (4.3)
Normal	31 (27.0)
Overweight	35 (30.4)
Obese	44 (38.3)
WC	
Normal	35 (30.4)
Central obesity	80 (69.6)
History of hormonal contraceptives	
History of hormonal contraceptives used (-)	40 (34.8)
History of hormonal contraceptives used (+)	75 (65.2)
Type of hormonal contraceptives	
Pill	8 (10.6)
Combination pill and injection	8 (10.6)
Injection	58 (75)
Implant	1 (1.3)

WCs: Waist circumferences

CONCLUSION

This study concludes that Minangkabau premenopausal women have shorter telomeres than those found in other samples. There is no significant correlation between BMI, WC, and estradiol level with telomere length. However, in people who have no history of hormonal contraceptive used, there is a significant correlation between estradiol levels and telomere lengths.

SIGNIFICANCE STATEMENT

This study demonstrates that the history of hormonal contraceptive use affects estradiol levels and telomere length.

ACKNOWLEDGMENT

We are grateful to Andalas University for funding this research project (Grant no 33/UN.16/HKRGB/LPPM/2017) .

CONFLICTS OF INTEREST

The authors have no conflicts of interests in this study.

AUTHOR'S CONTRIBUTION

Yusrawati, Delmi Sulastri, and Desmawati have a contribution in this publication. Yusrawati is the main author who has an idea to write about this and is a manuscript drafter. Delmi Sulastri helped correct the manuscript and was chairman of the study. Desmawati is a corresponding author, a member of the researcher team, manuscript drafter, and corrects the manuscript.

Table 3: Spearman's Rank correlation test result

Variables	Telomere length								
	All subject			History of hormonal contraceptives used (-)			History of hormonal contraceptives used (+)		
	n	r	p	n	r	p	n	r	p
BMI	115	0.064	0.500	40	−0.117	0.471	75	0.148	0.205
WC	115	0.39	0.682	40	−0.070	0.668	75	0.100	0.391
Estradiol levels	115	0.176	0.061	40	0.353	0.025*	75	0.113	0.334

*Correlation is significant at the 0.05 level (2-tailed)

REFERENCES

1. BKKBN. Laporan Kerja Instansi Pemerintah 2015. Jakarta: Badan Kependudukan dan Keluarga Berencana Nasional; 2016.
2. Fleischman DS, Navarrete CD, Fessler DM. Oral contraceptives suppress ovarian hormone production. Psychol Sci 2010;21:750-2.
3. Barrett EL, Richardson DS. Sex differences in telomeres and lifespan. Aging Cell 2011;10:913-21.
4. MFMER. Rochester Interpretive Handbook. Rochester, MN: Mayo Foundation for Medical Education and Research: Mayo Clinic, Mayo Medical Laboratories; 2016.
5. Shin YA, Lee KY. Low estrogen levels and obesity are associated with shorter telomere lengths in pre- and postmenopausal women. J Exerc Rehabil 2016;12:238-46.
6. Lin J, Kroenke CH, Epel E, Kenna HA, Wolkowitz OM, Blackburn E, *et al.* Greater endogenous estrogen exposure is associated with the telomere maintenance in postmenopausal women at risk for cognitive decline. Brain Res 2011;1379:224-31.
7. Ren F, Li C, Xi H, Wen Y, Huang K. Estimation of human age according to telomere shortening in peripheral blood leukocytes of tibetan. Am J Forensic Med Pathol 2009;30:252-5.
8. Dalgård C, Benetos A, Verhulst S, Labat C, Kark JD, Christensen K, *et al.* Leukocyte telomere length dynamics in women and men: Menopause vs age effects. Int J Epidemiol 2015;44:1688-95.
9. O'Callaghan NJ, Fenech M. A quantitative PCR method for measuring absolute telomere length. Biol Proced Online 2011;13:3.
10. Changamma C, Lalithamma A. The effect of estradiol valerate on sex hormone levels in aged female albino rats. Int J Curr Pharm Res 2015;7:36-8.
11. Ismail MY. Drug-food interactions and role of pharmacist. Asian J Pharm Clin Res 2009;2:1-10.
12. Boccardi V, Paolisso G. Malleability of short telomeres by telomerase activators: A mini-review. Aging Sci 2013;1:108.
13. Montpetit AJ, Alhareeri AA, Montpetit M, Starkweather AR, Elmore LW, Filler K, *et al.* Telomere length: A review of methods for measurement. Nurs Res 2014;63:289-99.
14. Gu Y, Honig LS, Schupf N, Lee JH, Luchsinger JA, Stern Y, *et al.* Mediterranean diet and leukocyte telomere length in a multi-ethnic elderly population. Age (Dordr) 2015;37:24.
15. Tiainen AM, Männistö S, Blomstedt PA, Moltchanova E, Perälä MM, Kaartinen NE, *et al.* Leukocyte telomere length and its relation to food and nutrient intake in an elderly population. Eur J Clin Nutr 2012;66:1290-4.
16. Harte AL, da Silva NF, Miller MA, Cappuccio FP, Kelly A, O'Hare JP, *et al.* Telomere length attrition, a marker of biological senescence, is inversely correlated with triglycerides and cholesterol in south asian males with Type 2 diabetes mellitus. Exp Diabetes Res 2012;2012:895185.
17. Furukawa S, Fujita T, Shimabukuro M, Iwaki M, Yamada Y, Nakajima Y, *et al.* Increased oxidative stress in obesity and its impact on metabolic syndrome. J Clin Invest 2004;114:1752-61.
18. Song Z, von Figura G, Liu Y, Kraus JM, Torrice C, Dillon P, *et al.* Lifestyle impacts on the aging-associated expression of biomarkers of DNA damage and telomere dysfunction in human blood. Aging Cell 2010;9:607-15.
19. Lindh I, Ellström AA, Milsom I. The long-term influence of combined oral contraceptives on body weight. Hum Reprod 2011;26:1917-24.
20. Atthobari J, Gansevoort RT, Visser ST, de Jong PE, de Jong-van den Berg LT, PREVEND Study Group, *et al.* The impact of hormonal contraceptives on blood pressure, urinary albumin excretion and glomerular filtration rate. Br J Clin Pharmacol 2007;63:224-31.
21. Shukla A, Jamwal R, Bala K. Adverse effect of combines oral contraceptive pills. Asian J Pharm Clin Res 2017;10:17-21.
22. Sulastri D, Lestari Y, Desmawati A. Relationship between body composition and smoking habit with telomere length of minangkabau ethnicity men, in West Sumatera, Indonesia. Pak J Biol Sci 2017;20:516-22.
23. Wright YL. Secret About Bioidentical Hormone: To Lost Fat and Prevent Cancer, Heart Disease, Menopause, Andropause By Optimizing Adrenals, Thyroid, Estrogen, Progesterone, Testosterone and Growth Hormone. USA: Lulu.com; 2011. p. 119.
24. Liu JP, Li H. Telomerase in the ovary. Reprod Fert 2010;140:215-22.
25. Bayne S, Jones ME, Li H, Pinto AR, Simpson ER, Liu JP. Estrogen deficiency leads to telomerase inhibition, telomere shortening and reduced cell proliferation in the adrenal gland of mice. Cell Res 2008;18:1141-50.
26. Breu A, Sprinzing B, Merkl K, Bechmann V, Kujat R, Jenei-Lanzl Z, *et al.* Estrogen reduces cellular aging in human mesenchymal stem cells and chondrocytes. J Orthop Res 2011;29:1563-71.