The effects of Korean red ginseng on biological aging and oxidative stress in postmenopausal women: A double-blind randomized controlled study

Tae-ha Chung

Department of Medicine

The Graduate School, Yonsei University

The effects of Korean red ginseng on biological aging and oxidative stress in postmenopausal women: A double-blind randomized controlled study

Tae-ha Chung

Department of Medicine

The Graduate School, Yonsei University

The effects of Korean red ginseng on biological aging and oxidative stress in postmenopausal women: A double-blind randomized controlled study

Directed by Professor Yong-jae Lee

The Doctoral Dissertation
submitted to the Department of Medicine
the Graduate School of Yonsei University
in partial fulfillment of the requirements for the degree
of Doctor of Philosophy

Tae-ha Chung

This certifies that the Doctoral Dissertation of Tae-ha Chung is approved.

Thesis Supervisor: Yong-jae Lee

Thesis Committee Member #1: Yong-ho Lee

Thesis Committee Member #2: Jong-koo Kim

Thesis Committee Member #3: Lark-kyun Kim

Thesis Committee Member #4: Chan-joo Lee

The Graduate School
Yonsei University

ACKNOWLEDGEMENTS

I would first like to thank Professor Yong-jae Lee for all of his devoted guidance, mentorship, and advice. I faced many difficulties in completing this study, but Professor Lee's sincere encouragement and love for me gave me the strength to endure to the end. He always told me to not be afraid to stand on the border. His teachings and trust in me were a constant source of growth and inspiration to try new things.

In addition, I would like to express my appreciation for having had the opportunity to work with Professors Jong-koo Kim, Yong-ho Lee, Chan-joo Lee, and Lark-kyun Kim. Their sincere guidance had a significant influence on me. Also, I would like to thank the many people in the Severance checkup, laboratory, and Department of Family Medicine, Severance hospital for their help in completing this study.

I dedicate this manuscript to my most beloved wife, Hye-jeong, lovely twins In-hoo and Ji-hoo, and parents, Ki-yong and Yoon-sook, who have always believed in and supported me.

<TABLE OF CONTENTS>

LIST OF FIGURES

LIST OF TABLES

ABSTRACT

The effects of Korean red ginseng on biological aging and oxidative stress in postmenopausal women: A double-blind randomized controlled study

Tae-ha Chung

Department of Medicine
The Graduate School, Yonsei University

(Directed by Professor Yong-jae Lee)

Background: Postmenopausal women are vulnerable to aging and oxidative stress due to reduced estrogen levels. Previous studies have shown that Korean red ginseng (KRG) has beneficial effects on aging and antioxidant capacity. Therefore, we evaluated the effects of the KRG on biological aging, oxidative stress, and antioxidant capacity in postmenopausal women.

Methods: This study conducted a double-blinded, placebo-controlled clinical trial. The participants were administered KRG or placebo, and the following metrics were measured: mitochondria DNA (mtDNA) copy number and DNA telomere length as indicators of biological aging, total oxidant status and total antioxidant status as markers of oxidative stress, and

antioxidant capacity. Clinical symptoms of fatigue, as measured by the fatigue severity scale, and menopause, as measured by the menopause rating scale, were measured before and after KRG administration.

Results: There were 63 participants, of whom 33 received KRG and 30 received a placebo. mtDNA copy number (KRG group: 1.58 ± 2.05, placebo group: 0.28 ± 2.36, p = 0.023) and TAS (KRG group: 0.11 ± 0.25 mmol/L, placebo group: -0.04 ± 0.16 mmol/L, p = 0.011) increased and fatigue severity score (KRG group: -7 ± 12, placebo group: -1 ± 11, p = 0.033) decreased significantly more in the KRG group than the placebo group.

Conclusion: KRG significantly increased the mtDNA copy number, total antioxidant status, and improved symptoms of fatigue in postmenopausal women.

Keywords: korean red ginseng, oxidative stress, antioxidant activity, biological aging, fatigue, postmenopausal women

The effects of Korean red ginseng on biological aging and oxidative stress in postmenopausal women: A double-blind randomized controlled study

Tae-ha Chung

Department of Medicine
The Graduate School, Yonsei University

(Directed by Professor Yong-jae Lee)

I. INTRODUCTION

Ginseng was traditionally used in East Asian countries, including Korea, China, and Japan, as a functional health food and natural therapeutic medicine, but it is increasingly being recognized as a natural health food internationally.[1, 2] Korean red ginseng (KRG), the technical name of which is *Panax ginseng* Meyer, is produced by steaming and drying fresh, unpeeled ginseng. KRG has anti-cancer and antioxidant properties and consuming it improves immune system activity, fatigue systems, blood circulation, and memory function.[2]

Although no exact mechanism has been identified for the aging process in women after menopause, several epidemiological studies have shown that there are

many diseases related to aging in postmenopausal women. Levine et al. showed that an aging biomarker that was identified via DNA methylation to determine epigenetic age increased in the blood over time after menopause began.[3] In a prospective study of 12,134 Dutch women, age-adjusted mortality increased by 2% in women who began menopause more than a year earlier.[4] In a Korean study, Kim et al. analyzed data from the Korean National Health and Nutrition Examination Survey and found that postmenopausal women were 1.6 times more likely to develop a metabolic syndrome than premenopausal women.[5] A large cohort study on women, including postmenopausal women, showed that various metabolic parameters associated with type 2 diabetes and cardiovascular disease rates, both of which are closely linked to oxidative stress and chronic low-grade inflammation, dramatically increased after menopause began.[6]

Telomeres are nucleoprotein structures at the terminal end of linear chromosomes that protect the tails of the linear chromosomes from degeneration and maintain overall stability.[7] During cell division, telomeres become shorter and eventually induce cell senescence and apoptosis.[8] Mitochondria dysfunction is associated with senescence.[9] Both mtDNA mutations and decreases in mtDNA copy number interrupt mitochondrial function, which may lead to aging.[8, 10] Healthy adults and men with metabolic syndromes who consumed KRG had decreased the damage of DNA tail lengths, which is associated with biological aging, and increased mtDNA copy number.[11-13]

Free radicals accumulate in macromolecules, such as fat, DNA, and protein. Moreover, oxidative stress leads to cellular senescence, which induces the expression of various inflammatory cytokines and proteins, which in turn promote aging.[14, 15] The

previous clinical studies of the antioxidant effects of the KRG showed that the antioxidant indicators such as superoxide dismutase (SOD), glutathione peroxidase (GPx), and catalase increased, whereas malondialdehyde (MDA) as an oxidative stress maker decreased in postmenopausal women.[12] However, the measurement of these oxidative stress and antioxidant indicators have limitations in primary clinical settings. As the process of examination is complex and mainly laboratory-measurable, it cannot easily be used in clinical practice. Meanwhile, a colorimetry diagnosis technique has recently been developed for the assessment of oxidative stress and antioxidative activity using simple blood tests in clinical practice. This method evaluates all oxidative stress and antioxidative indicators, including both enzymatic and non-enzymatic molecules in the blood, and yields total oxidant status (TOS) and total antioxidant status (TAS).[16, 17]

Unlike previous studies, both DNA telomere length and mtDNA copy number of lymphocytes were used to determine biological aging, TOS and TAS were measured to evaluate oxidative stress and antioxidant capability with diagnostic indicators that can easily verify in the primary clinical setting in this study. Furthermore, this study used the Fatigue Severity Scale (FSS) and Menopause Rating Scale (MRS) questionnaires as indicators of clinical symptoms for fatigue and postmenopausal symptoms.

Estrogen directly protects against mitochondria apoptosis and reduces oxidative stress.[18] Thus, this study was conducted with postmenopausal women whose biological aging would be faster and antioxidant activity would be lower than premenopausal women because they have low estrogen. In addition, postmenopausal women were selected as the study subjects to control the effects of lifestyle habits such as smoking and drinking, which are relatively more linked to aging and oxidative stress

than men. This study was conducted primarily to evaluate how KRG affected biological aging, oxidative stress, and antioxidant status, and secondarily to evaluate how it affected clinical symptoms.

II. METHODS AND MATERIALS

1. Study design and participants

This study comprised an 8-week, randomized, double-blind, placebo-controlled, clinical trial. Participants were examined at the Severance Health Check-up, Severance Hospital, Yonsei University Health System, in Seoul, Korea, between August 2019 and March 2021. This study's protocol was approved by the Institutional Review Board of Gangnam Severance Hospital (IRB no.: 3-2019-0089) and registered at the Clinical Research Information Service (CRIS, KCT0006506). This study was performed in compliance with the Declaration of Helsinki.

Eligible participants were postmenopausal women aged 46–69 years who could communicate in Korean naturally and who were not participating in any other interventional studies. Additionally, participants had to have no physical restrictions on movement. Those who were excluded were current smokers; those who consumed more than 3 standard alcoholic drinks more than two days per week; who had uncontrolled hypertension, defined as systolic blood pressure (SBP) ≥ 180 mmHg or diastolic blood pressure (DBP) ≥ 100 mmHg; who had uncontrolled diabetes mellitus, defined as having fasting plasma glucose ≥ 200 mg/dL; who had been diagnosed with liver disease and aspartate aminotransferase (AST) or alanine aminotransferase (ALT) levels more than three times the upper limit; who had been diagnosed with kidney, cardiovascular,

or cerebral vascular disease; who had any form of cancer or were undergoing cancer treatment; or who were taking other health supplements or female hormones. Written informed consent was obtained from all patients before participation.

A total of 73 participants were enrolled in this study, 37 of whom were randomly placed in the KRG group and 36 of whom were randomly placed in the placebo group. During the study, 4 participants from the KRG group were excluded, 2 of whom stopped participating for unknown reasons, 1 of whom was hospitalized to receive surgery, and 1 of whom took multivitamin supplements. Six participants were excluded from the placebo group, 1 of whom had abdominal pain, 2 of whom had constipation, 1 of whom took multivitamin supplements, and 2 of whom took other red ginseng products. A total of 33 participants in the KRG group and 30 participants in the placebo group completed the study. Figure 1 shows a flow chart for how the study was conducted.

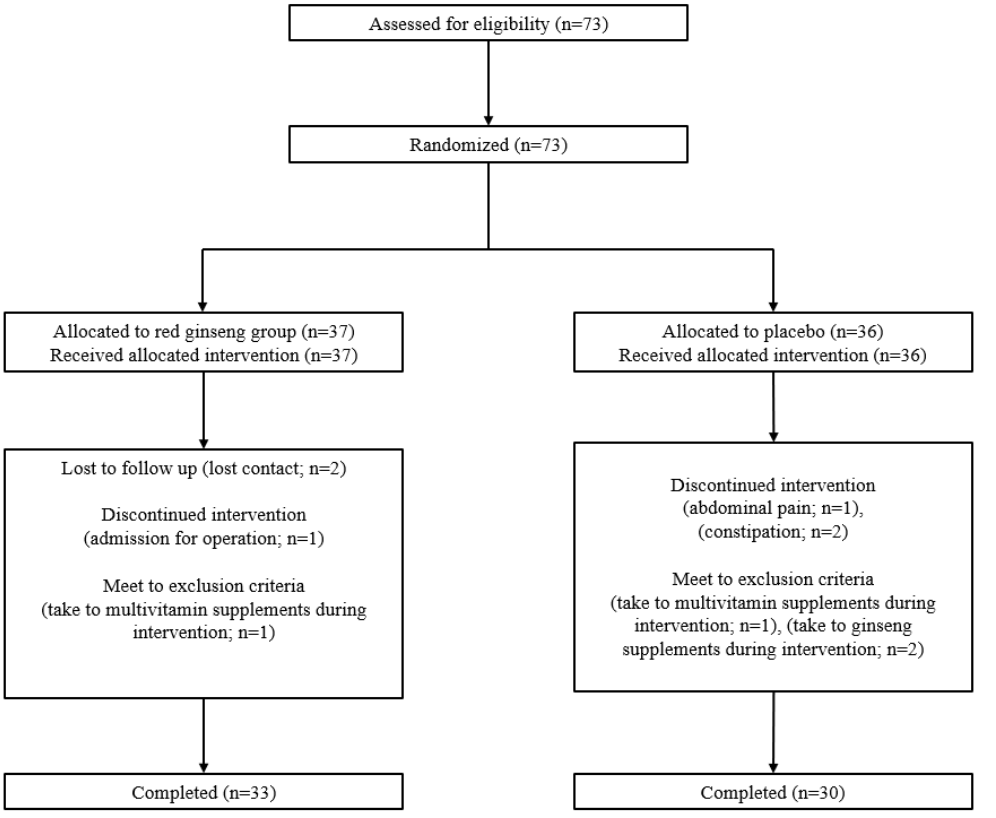


Figure 1. Study procedure flowchart

2. Randomization

Participants were randomly assigned to take a 500 mg KRG or placebo tablet four times daily. Participants were assigned to each group in an even ratio by sequentially assigning them in alternating order by their assignment code. These codes were generated using SAS version 9.4 (SAS Institute, Inc., Cary, NC, USA). Both researchers and participants were blinded to the assignments throughout the study.

3. Korean red ginseng and placebo supplements

Participants in the KRG group received 2 g of KRG tablet per day. The tablets contained 8.03 mg/g of ginsenoside Rb1, 3.29 mg/g of Rc, 2.80 mg/g of Rb2, 2.50 mg/g of Rg3, 1.47 mg/g of Rf, 1.29 mg/g of Re, 1.18 mg/g of Rg1, and 1.0 mg/g of Rd. The KRG tablets were prepared by dehydrating KRG extract (3 g of KRG extract per 2 g tablet). The placebo tablets contained cornstarch and cellulose with the same flavor as the KRG tablets. The KRG and placebo tablet were provided by the study sponsor (The Korean Society of Ginseng, Seoul, Korea).

4. Data collection

Each participant completed a questionnaire about whether they smoked cigarettes, whether they consumed more than 3 standard alcoholic drinks on more than 2 days per week, whether they engaged in moderate-to-vigorous intensity physical activity more than 3 times per week, their medical history, and whether they consumed health supplements. Body mass and height were measured to the nearest 0.1 kg and 0.1 cm, respectively, while participants wore light indoor clothing and no shoes. Body mass index was defined as the participant's weight in kilograms divided by the square of their height in meters. SBP and DBP were measured using the patient's right arm with a standard mercury sphygmomanometer (Kensei Industry Co., Ltd., Takasai Shimotsuma, Japan). Participants were defined as having hypertension if their SBP ≥ 140 mmHg, their DBP ≥ 90mmHg, they were taking anti-hypertension medication, or had been diagnosed with hypertension by a physician. Participants were defined as having diabetes if their fasting plasma glucose ≥ 126 mg/dL, their glycated hemoglobin

level ≥ 6.5%, they were taking anti-diabetes medication, or had been diagnosed with diabetes by a physician. Participants were defined as having dyslipidemia if their total cholesterol ≥ 240mg/dL, low-density lipoprotein (LDL) cholesterol ≥ 160mg/dL, triglycerides ≥ 150mg/dL, or high-density lipoprotein (HDL) cholesterol < 50mg/dL, they were taking anti-dyslipidemia medication, or had been diagnosed with dyslipidemia by a physician. Participants were defined as being postmenopausal if they had not had a menstrual period for at least the preceding 12 months in the absence of a clear biological or physiological cause.

Blood samples were obtained after 8 hours of overnight fasting. Leukocyte counts were measured using the ADVIA 2120i Hematology System (Siemens Healthcare Diagnostic, Inc., NY, USA). Fasting plasma glucose, total cholesterol, triglycerides, LDL cholesterol, HDL cholesterol, AST, ALT, and gamma-glutamyltransferase (gamma-GT) were measured with an ADVIA 1800 Clinical Chemistry System (Siemens Healthcare Diagnostic, Inc., NY, USA).

5. Dependent variable assessment

A. DNA telomere length

We measured the DNA telomere length of lymphocytes by performing monochrome multiplex quantitative polymerase chain reactions (PCR) on the participants' whole blood samples.[19] Telomere length quantitative real-time PCR was performed with a Bio-Rad MyiQ Single Color Real-Time PCR Detection System. For multiplex quantitative real-time PCR, the telomere primer pair telg and gelc with final concentrations of 900 nM each was combined with either the albumin primer pair albu

and albd with final concentrations of 900 nM each or with the beta-globin primer pair hbgu and hbgd with final concentrations of 500 nM each in the master mix. The PCR cycles for monochrome multiplex quantitative PCR were as follows: stage 1 was 15 min at 95℃; stage 2 was 2 cycles of 15s at 94℃ and 15s at 49℃; and stage 3 32 cycles of 15s at 94℃, 10s at 62℃, 15s at 74℃ with signal acquisition for telomere amplification, 10s at 84℃, and 15s at 88℃ with signal acquisition for scg amplification. After thermal cycling and data collection were complete, the MyiQ Bio-Rad iQ5 2.0 Standard Edition Optical System Software (Bio-Rad Laboratories, Inc., CA, USA) was used to generate two standard curves for the telomere and scg signals. Each DNA standard curve was generated using a genomic DNA sample and was used to calculate the ratio of telomere signals to scg signals.

B. Total oxidant status and total antioxidant status

TOS and TAS were measured using a TBA c8000 automated colorimeter (Toshiba Medical system Ltd, Tokyo, Japan). The TOS assay was conducted based on the oxidation of the ferrous ion-chelator complexes to ferric ions in the presence of different reactive oxidant species in an acidic medium. The oxidation ratio was calculated using xylenol orange. Ferric ions present a colored complex when they react with chromogen in a reagent medium (Rel Assay Diagnostics, Gaziantep, Turkey). Color intensity is linked to the total amount of oxidant molecules in the sample. The assay was conducted with hydrogen peroxide.[17]

The antioxidative molecules in the sample reduce dark blue-green-colored 2,2'-azinobis(3-ethylbenzo-thiazoline-6-sulfonate) radicals into a colorless form

according to antioxidant concentrations and capacities. The change in absorbance at 660 nm is related to the total amount of antioxidants in the sample. The TAS assay was calibrated with Trolox equivalent, a stable, standard antioxidant solution and a vitamin E analog.[16]

C. Mitochondria DNA copy number

All DNA samples were extracted from ethylene-diamine-tetraacetic acid (EDTA) blood. High-quality genomic DNA was extracted from participants' peripheral blood using a QIAamp DNA Mini Kit (Qiagen, Valencia, CA, USA) according to the manufacturer's protocol. The concentration and purity of total DNA were controlled by taking absorbance readings at 260 nm and 280 nm using a Nanodrop 1000 spectrophotometer (preQLab Biotechnology, Erlangen, Germany). The relative mtDNA copy number was measured by quantitative real-time PCR. Two pairs of primers were designed and used to quantify the mtDNA copy number. The first primer pair was used to amplify the ND5 gene in the mtDNA. The forward primer (ND5-F) was 5'-TTCATCCCTGTAGCATTGTTCG-3' and the reverse primer (ND5-R) was 5'-GTTGGAATAGGTTGTTAGCGGTA-3'. The second primer pair was used to amplify the nuclear DNA rRNA 18S. The forward primer (18S-F) was 5'-GAGAAACGGCTACCACATCC-3' and the reverse primer (18S-R) was 5'-GCCTCGAAAGAGTCCTGTATTG-3'. Ten uL of the PCR reaction mixture for mtDNA amplification consisted of 2 X SYBR Green Mastermix (Applied Biosystems, MA, USA), 100 pmol/uL of ND5-F or 18S-F primer, 100 pmol/uL of ND5-R or 18S-R primer, and 10 ng of genomic DNA. The samples were heated to 95℃ for 10 min followed by 40 cycles of heating to 95℃ for 15 s and then cooling to 55℃ for 1 min.

Each sample was run in triplicate in a 96-well plate with an Applied Biosystems 7300 real-time PCR system (Applied Biosystems, MA, USA). The relative copy number was calculated from the threshold cycle value (ΔCt), which was defined as Ct_{18S} - Ct_{ND5}, where the mean amount of mtDNA = $2^{-\Delta Ct}$. The fold change of the mtDNA copy number was calculated as the amount of ND5, the target gene, DNA normalized to the amount of 18S, the reference gene, DNA in a treated sample.

D. Fatigue Severity Scale

The FSS is a popular tool for measuring fatigue symptoms.[20] It has been validated for various diseases, such as multiple sclerosis, systemic lupus erythematosus, Parkinson's disease, and stroke.[21-23] It consists of 9 items, each of which can receive a score of 1–7 points. Fatigue severity is the sum of each item's score for a total score range of 7–63 where higher scores indicate more severe fatigue symptoms (Appendix 1).

E. Menopause Rating Scale

The Menopause Rating Scale (MRS) measures the severity of aging symptoms in women.[24] It is widely used in primary clinical practice and has been validated.[25] It consists of 4 psychological items, 4 somatic items, and 3 urogenetic items. Each item can receive a score of 0–4 points for a total score range of 0–44 with higher scores indicating more severe symptoms (Appendix 2).

6. Statistical analyses

We analyzed the data based on the law of large numbers and the central limit theorem because each group had at least 30 participants. The groups' baseline characteristics were compared using independent t-tests for continuous variables and chi-squared tests for categorical values. We used a paired t-test to determine whether the groups' metabolic parameters differed after the intervention. The mean differences in changes between the groups were compared using independent two-sample t-tests. And, we used a Wilcoxon signed-rank test and Mann Whitney U test in subgroup analysis. All statistical analyses were performed using SPSS version 25.0 for Windows (IBM, NY, USA). All statistical tests were two-sided and *P*-values < 0.05 were considered statistically significant.

III. RESULTS

The groups' baseline characteristics are presented in Table 1. The groups had similar anthropometric parameters. The average age of the KRG group was 58.7 ± 4.2 years old and that of the placebo group was 59.7 ± 4.2 years old. There were no significant differences between the two groups' results in terms of SBP, DBP, waist circumferences, body mass index, fasting plasma glucose levels, triglycerides, HDL cholesterol, LDL cholesterol, AST, ALT, gamma-GT levels, telomere length, mtDNA copy number, TOS, TAS, FSS scores, MRS scores, alcohol consumption rates, physical activity rates, or prevalence of hypertension, diabetes, or dyslipidemia. However, the KRG group's average leukocyte count was 4,890 ± 931 cells/μL while that of the placebo group was 5,553 ± 1361 cells/μL ($p = 0.027$), and the KRG group's total cholesterol level was 230

± 31 mg/dL, while that of the placebo group was 210 ± 41 mg/dL (p = 0.034).

Table 1. Baseline characteristics of study participants

	KRG	Placebo	P-value
N	33	30	
Age, years	58.7±4.2	59.7±4.2	0.330
SBP (mmHg)	120.7±15.3	125.0±11.1	0.207
DBP (mmHg)	73.0±11.6	77.2±7.9	0.104
Waist circumference (cm)	79.3±6.9	80.9±7.2	0.371
BMI (kg/m^2)	23.7±2.4	23.8±3.1	0.844
Leukocyte count (cells/μL)	4890±931	5553±1361	0.027
Fasting plasma glucose (mg/dL)	95±11	96±17	0.831
Total cholesterol (mg/dL)	230±31	210±41	0.034
Triglyceride (mg/dL)	129±66	116±52	0.392
HDL cholesterol (mg/dL)	68±16	65±16	0.373
LDL cholesterol (mg/dL)	136±38	124±36	0.205
AST (U/L)	27±8	25±4	0.360
ALT (U/L)	23±9	23±7	0.873
Gamma-GT (U/L)	17±11	16±7	0.606
Alcohol consumption, n (%)	3 (9.1)	5 (16.7)	0.367
Physical activity, n(%)	17 (51.5)	10 (33.3)	0.145
HTN, n(%)	4 (12.1)	4 (13.3)	0.885
Diabetes, n(%)	1 (3.0)	2 (6.7)	0.498
Dyslipidemia,n (%)	5 (15.2)	7 (23.3)	0.409

TAS (mmol/L)	1.42±0.16	1.44±0.14	0.690
TOS (μmol/L)	3.64±1.18	3.74±1.04	0.704
ΔTelomere length (Kb)	-0.04±0.21	-0.04±0.27	0.964
mtDNA copy number	4.35±2.54	4.70±3.94	0.678
FSS	34±16	32±15	0.619
MRS	10±8	11±7	0.832

p-values were calculated using the independent two-sample
t-test for continuous values and the chi-square test for categorical value
BMI, body mass index; WC, waist circumference; SBP, systolic blood pressure; DBP, diastolic blood pressure; AST, aspartate aminotransferase; ALT, alanine aminotransferase; Gamma-GT, gamma-glutamyltransferase; HTN, hypertension; TAS, total anti-oxidative status; TOS, total oxidative status; ΔTelomere length= age-standard telomere length – currently measured telomere length; mtDNA copy number, mitochondria DNA copy number; FSS, fatigue severity scale; MRS, menopause rating scale

Table 2 presents the changes in the metabolic parameters in the two groups over the course of the study. Body mass index, SBP, DBP, and hematologic parameters did not change significantly in either group. The KRG group's LDL cholesterol tended to decrease from 136 ± 38 mg/dL to 127 ± 34 mg/dL compared to the placebo group (p=0.067). However, the two groups had no significant difference in mean changes of LDL cholesterol (p = 0.064).

Table 2. Changes in metabolic parameters of KRG and placebo before and after 8 weeks

	KRG			Placebo			
	Baseline	**8 weeks**	**P-value**	**Baseline**	**8 weeks**	**P-value**	**Changed P-value**
BMI (kg/m^2)	23.7±2.4	23.7±2.40	0.755	23.8±3.1	23.9±3.00	0.811	
Change	0±0.5			0.1±0.1			0.972
SBP (mmHg)	120.7±15.3	120.3±14.9	0.822	125.0±11.1	124.5±12.7	0.819	
Change	-0.4±9.9			-0.5±11			0.973
DBP (mmHg)	73.3±11.6	74.4±9.3	0.255	77.2±9.6	77.3±7.9	0.948	
Change	1.4±6.7			0.1±8			0.511
Leukocyte count (cells/μL)	4890±931	4886±1385	0.800	5553±1361	5354±1380	0.414	
Change	-4±970			-199±1317			0.505
Fasting plasma glucose (mg/dL)	95±11	95±8	0.969	96±17	96±16	0.736	
Change	0±9			0±8			0.847
Total cholesterol (mg/dL)	230±31	223±31	0.172	210±41	209±44	0.773	
Change	-7±30			-1±31			0.465
Triglyceride (mg/dL)	129±66	137±124	0.560	116±52	107±44	0.258	
Change	8±80			-9±41			0.305
HDL cholesterol (mg/dL)	68±16	67±16	0.549	65±16	63±15	0.240	
Change	-1±7			-2±8			0.619

LDL cholesterol (mg/dL)	136±38	127±34	0.067	124±36	128±42	0.414	
Change	-9±24			4±26			0.064
AST (U/L)	27±8	25±4	0.334	25±4	27±6	0.103	
Change	-2±8			2±6			0.082
ALT (U/L)	23±9	22±9	0.608	23±7	24±9	0.536	
Change	-1±7			1±10			0.414
Gamma-GT (U/L)	17±11	19±12	0.395	16±7	16±5	0.346	
Change	-2±7			0±4			0.243

p-values were calculated using the paired t-test (difference after intervention within groups), Changed p-values were calculated using the independent two-sample t-test (mean change difference between two groups).
BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; AST, aspartate aminotransferase; ALT, alanine aminotransferase; Gamma-GT, gamma-glutamyltransferase

Table 3 presents data regarding this study's primary variables of interest, including antioxidant capacity, oxidant and biological aging indicators. TAS increased significantly in the KRG group, from 1.42 ± 0.16 mmol/L to 1.53 ± 0.19 mmol/L (p = 0.021), but did not statistically significantly change in the placebo group, decreasing from 1.44 ± 0.14 mmol/L to 1.40 ± 0.17 mmol/L (p = 0.258). The KRG group's mean change in TAS of 0.11 ± 0.25 mmol/L was significantly greater than that of the placebo group at -0.04 ± 0.16 mmol/L (p = 0.011). The KRG group's TOS was significantly higher than baseline, increasing from 3.64 ± 1.18 μmol/L to 4.50 ± 1.80 μmol/L (p = 0.029), but the placebo group's was not, increasing from 3.75 ± 1.04 μmol/L to 4.38 ± 2.52 μmol/L (p = 0.196). The mean change in TOS was not significantly different between the two groups (p = 0.713).

The biological indicators, ΔTelomere length is a difference between age-

standard telomere length and currently measured participant`s telomere length. It did not significantly change for either group during the study (KRG group: p = 0.800, placebo group: p = 0.616). However, mtDNA copy number was significantly higher in the KRG group, increasing from 4.36 ± 2.54 to 5.93 ± 3.33 (p < 0.001), but not in the placebo group, increasing from 4.70 ± 3.94 to 4.98 ± 3.17 (p = 0.520). The mean change in mtDNA copy number was significantly different between the groups, with the KRG group's being 1.58 ± 2.05 and the placebo group's being 0.28 ± 2.36 (p = 0.023). Furthermore, the fold change refers to the multiple when the baseline mtDNA copy number is considered to be 1. The mtDNA copy number fold change in the KRG group was 1.44 ± 0.46, which was significantly higher than the placebo group 1.15 ± 0.40 (p = 0.012).

Table 3. Changes in Oxidative and biological aging indicators of KRG and placebo before and after 8 weeks

	KRG			Placebo			
	Baseline	**8 weeks**	**P-value**	**Baseline**	**8 weeks**	**P-value**	**Changed P-value**
TOS (μmol/L)	3.64±1.18	4.50±1.80	0.029	3.75±1.04	4.38±2.52	0.196	
Change	0.86±2.16			0.64±2.64			0.713
TAS (mmol/L)	1.42±0.16	1.53±0.19	0.021	1.44±0.14	1.40±0.17	0.258	
Change	0.11±0.25			-0.04±0.16			0.011

ΔTelomere length(Kb)	-0.04±0.21	-0.03±0.24	0.800	-0.04±0.27	-0.05±0.27	0.616	
Change	0.01±0.075			-0.01±0.061			0.606
mtDNA copy number	4.36±2.54	5.93±3.33	<0.001	4.70±3.94	4.98±3.17	0.520	
Change	1.58±2.05			0.28±2.36			0.023
Fold change	1.44±0.46			1.15±0.40			0.012

p-values were calculated using the paired t-test (difference after intervention within groups), Changed p-values were calculated using the independent two-sample t-test (mean change difference between two groups).
TAS, total anti-oxidative status; TOS, total oxidative status; mtDNA copy number, mitochondria DNA copy number; ΔTelomere length= age-standard telomere length−currently measured telomere length

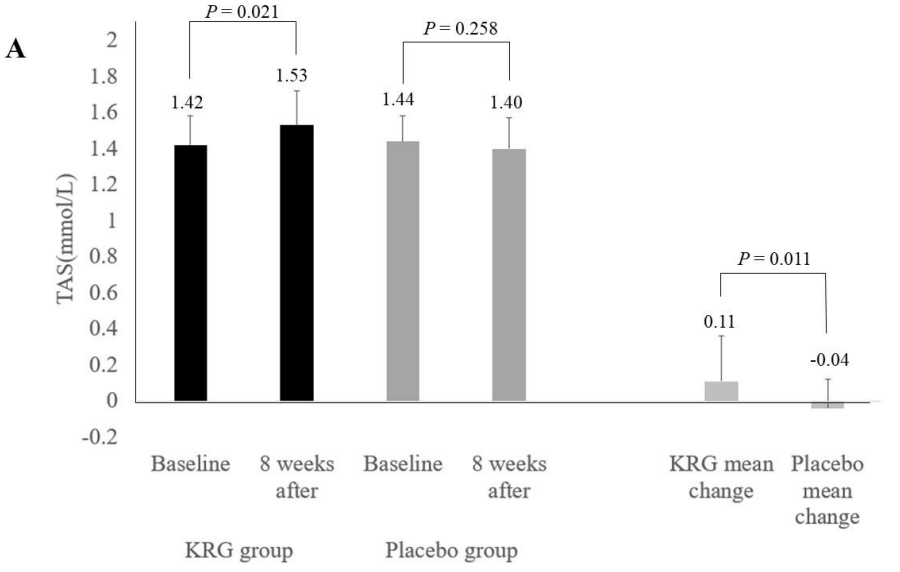
The results regarding fatigue and menopause symptoms are presented in Table 4. FSS scores decreased significantly in the KRG group from 34 ± 16 to 27 ± 14 (p = 0.002) but did not significantly change in the placebo group, decreasing from 32 ± 15 to 31 ± 16 (p = 0.861). The mean change in the KRG group's FSS scores was -7 ± 12, which was significantly different from the placebo group's -1 ± 11 (p = 0.033). However, the groups' MRS scores were not significantly different (KRG group: p = 1.000, placebo group: p = 0.058).

Table 4. Changes in questionnaire (FSS, MRS) of KRG and placebo before and after 8weeks

	KRG			Placebo			
	Baseline	**8 weeks**	**P-value**	**Baseline**	**8 weeks**	**P-value**	**Changed P-value**
FSS	34±16	27±14	0.002	32±15	31±16	0.861	
Change	-7±12			-1±11			0.033
MRS	10±8	10±9	1.000	11±7	8±7	0.058	
Change	0±5			-3±7			0.100

p-values were calculated using the paired t-test (difference after intervention within groups), Changed p-values were calculated using the independent two-sample t-test (mean change difference between two groups).
FSS, fatigue severity scale; MRS, menopause rating scale

Finally, the significant results of this study are summarized in Figure 2.



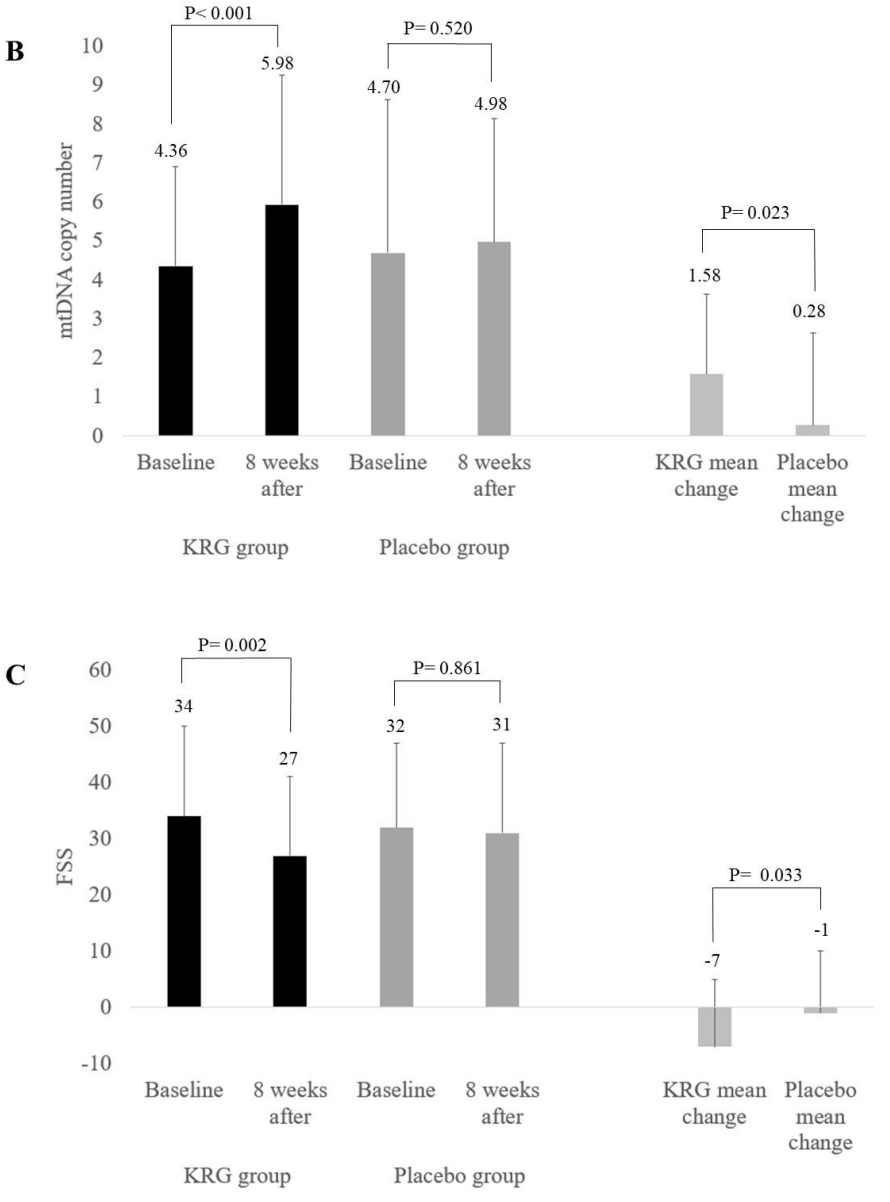


Figure 2. Changes in major results of KRG and placebo before and after 8 weeks (bars mean standard deviation); (A) Total antioxidant status, (B) mtDNA copy number, (C) Fatigue severity scale

Table 5 showed that the change in FSS according to the interval change of TAS and mtDNA copy number before and after intervention in the KRG group. The symptom of fatigue statically decreased associated with increased TAS and mtDNA copy number after intervention (p=0.011, p=0.003).

Table 5. The FSS of KRG group according to interval change of TAS and mtDNA copy number before and after 8weeks.

	Group 1†			Group 2‡			
	Baseline	**8 weeks**	**P-value**	**Baseline**	**8 weeks**	**P-value**	**Changed P-value**
FSS[1]	36 (27-48)	25 (16-45)	0.071	33 (17-46)	27 (12-33)	0.011	0.538
FSS[2]	34 (20-46)	27 (22-33)	0.345	34 (15-49)	26 (12-38)	0.003	0.833

Data are presented as median (interquartile ranges).
p-values were calculated using the Wilcoxon signed-rank test (difference after intervention within groups), Changed p-values were calculated using the Mann Whitney U test (change difference between two groups).
[1] FSS in the TAS
[2] FSS in the mtDNA copy number
†Groups 1 is defined as no interval change or decreased the TAS and the mtDNA copy number after intervention.
‡Groups 2 is defined as increased the TAS and the mtDNA copy number after intervention.
KRG, Korean red ginseng; TAS, total antioxidant status; mtDNA copy number, mitochondria DNA copy number; FSS, fatigue severity scale

IV. DISCUSSION

This study comprised a randomized, double-blind, placebo-controlled clinical trial to evaluate the effect of KRG consumption on biological aging, oxidative stress, antioxidant capacity, and fatigue and menopause symptoms in postmenopausal women. We found that taking 2g/day of KRG for 8 weeks increased mtDNA copy number and TAS and reduced fatigue symptoms more than the placebo.

Some of our findings were consistent with those of previous studies on the effectiveness of KRG while others were not. First, in this study, KRG was shown to increase mtDNA copy number more than the placebo. A previous study showed that KRG improved exercise performance, increased the ATP production capacity of muscle cells, and stimulated myoblast differentiation in mice.[26] An animal study showed that the mtDNA copy number and messenger RNA expression levels of mitochondrial biogenesis-related transcription factors increased more in those with diabetes who took KRG than those who took a placebo group.[27] A clinical trial showed that the mtDNA copy number for men with metabolic syndromes increased when they consumed 3g/day of KRG and the mean change in their mtDNA copy number was significantly higher than the group who consumed a placebo.[13]

The following mechanism can be considered. Peroxisome proliferator-activated receptor-gamma co-activator 1α (PGC-1α) plays a pivotal role in the regulation of mitochondrial biogenesis. It activates nuclear respiratory factor (NRF)-1 and NRF-2, thereby promoting the expression of mitochondrial transcription factor A (TFAM), which upregulates mitochondrial DNA transcription and replication.[28, 29] Moreover, Kim et al. reported that the Rg3 component in KRG increased the expression of PGC-1α, NRF-1, and TFAM in an in vitro model.[30] Additionally, Shin et al. showed that KRG improved swimming performance in mice and enhanced TFAM and NRF-1 gene expression levels.[26] Therefore, KRG may improve the PGC-1α-NRF1-TFAM pathway, which increases the mtDNA copy number. The mtDNA copy number is associated with aging and oxidative stress.[9, 31] Finally, our result suggests that consuming KRG can increase mitochondrial functioning, which is negatively associated with biological aging.

Second, the KRG had significantly higher TAS levels following the intervention than the placebo group. Studies on the antioxidant activity of KRG in animal models have shown that the levels of antioxidant indicators, such as SOD, GPx, and catalase, increased and the levels of the oxidative stress marker MDA decreased.[32-34] Seo et al. showed that postmenopausal women who took 3 g/day of KRG for 12 weeks had significantly higher SOD levels than those who took a placebo.[12] Another clinical trial in Korea showed that healthy adults who took 6 g/day of KRG had greater increases in GPx, SOD, and catalase activity than those who took a placebo.[11] Our results were consistent with these studies. However, a significant difference between this study and those was that antioxidant indicators, which can easily be detected in primary care settings, were used in this study rather than markers like SOD, GPx, and catalase that are much harder to detect in clinical settings. And, even if the dose and treatment duration of KRG were lower than in previous studies, it had an effect on antioxidant capacity. Although the antioxidant mechanism of KRG is not clearly understood, the following mechanism can be considered. First of all, it may improve antioxidant capacity by downregulating reactive oxygen species (ROS)-stimulated mitogen-activated protein kinase and protein kinase B pathways that are involved in cell survival and growth.[35, 36] Furthermore, KRG can upregulate nuclear factor erythroid 2-related factor 2 (Nrf 2)-mediated expression of heme oxygenase 1 as a ROS scavenging factor.[37] Additionally, the intracellular concentration of glutathione (GSH) was increased and attenuated the reduction of GSH in the cell levels.[38]

Third, TOS, which is an indicator of oxidative stress, increased in both groups, but only the KRG group's change was statistically significant. However, the difference of mean change TOS levels between the two groups was not statically significant.

Based on these results, we were carefully thought that some confounding factors, such as diet, exercise, physical or psychogenic stress, could make the KRG group more susceptible to oxidative stress during the intervention. Eventually, oxidative stress indicators increased in both groups, especially in the KRG group, but there was no statistically significant difference between the two groups. It confirmed that the KRG helped in increasing antioxidant capacity in this study.

Fourth, the KRG group showed improvement in fatigue symptoms. A meta-analysis reported that red ginseng intake does not statistically significantly reduce fatigue symptoms.[39] A randomized, placebo-controlled study did not show that KRG consumption resulted in a significant decrease in fatigue symptoms for the full sample, but a subgroup analysis showed that it did for people over 50 years old with moderate fatigue symptoms.[40] In this study, the KRG could reduce fatigue symptoms in postmenopausal women by confirming a statically significant after the intervention. Furthermore, we have performed a subgroup analysis to investigate the association of changes in fatigue symptoms with TAS and mtDNA copy number. This analysis confirmed that reducing fatigue symptoms in the KRG group was associated with increasing TAS and mtDNA copy number. Therefore, taking 2g of KRG per day for 8 weeks increased TAS and mtDNA copy number, which could help improve fatigue symptoms.

Fifth, telomere length, a biological aging indicator, did not statistically significantly change for either group and there was no statistically significant difference in the groups' mean changes in telomere length. Previous studies using animal models showed that *Panax ginseng* extract protects DNA by reducing cytochrome P450 (CYP)1A1 expression and oxidative stress.[41, 42] Although our results showed that KRG

did not affect telomere length, Kim et al. reported that DNA damage tail length and tail moment decreased according to KRG dose[11]. However, the results between this study and that study may have been different because KRG was administered at 3 and 6 g/day and the participants were adults with an average age of 36 years old. In addition, it thought that there was a difference in the results because the DNA damage tail length was measured rather than the direct DNA telomere length. In this study, if the KRG dose had been higher, the change in telomere length may have been significantly higher, so further research is necessary.

Mean LDL cholesterol levels in the KRG group decreased from 136 to 127 mg/dL, but they did not decrease in the placebo group. However, this decrease was not statistically significant ($p = 0.067$). A previous study showed that administering 6 g/day of *Panax ginseng* extract for 8 weeks decreased LDL cholesterol in males.[43] Another clinical study with hyperlipidemic patients reported that LDL cholesterol decreased significantly in the red ginseng (2g/day) and placebo group, but there was no significant difference between the two groups.[44] The subjects of that study were young, male, and hyperlipidemic, while this study was conducted on postmenopausal women. Therefore, this difference seems to indicate a dissimilarity in the study results. However, even though the baseline LDL cholesterol level was in the normal range, it showed a tendency to decrease after the intervention in postmenopausal women. A large sample and long-term clinical study are necessary to evaluate the effect of the KRG on lipid metabolism in the future.

This study has several strengths. Most related studies used SOD, GPx, MDA, and catalase as indicators of oxidative stress and antioxidant capacity, but these markers are difficult to detect in general clinical settings. We confirmed that the antioxidant

ability was increased in the KRG group by quantitatively measuring the total oxidative stress and various factors involved in antioxidant capacity in each individual's blood sample, differently from the previous methods. Furthermore, telomere length was measured directly to evaluate the biological aging, but it did not statistically significantly differ between the groups. However, this study showed that mtDNA copy number, which is related to mitochondrial function, energy metabolism, and the aging process, was enhanced, indicating that the KRG might protect against aging and increase cellular metabolism. In addition, we objectively evaluated fatigue and postmenopausal symptoms through validated questionnaires and showed that the KRG group's fatigue symptoms decreased. Finally, estrogen directly protects against mitochondrial apoptosis and reduces oxidative stress.[18] Thus, this study was conducted with postmenopausal women whose biological aging would be faster and whose antioxidant activity would be lower than that of premenopausal women because they have low estrogen. Furthermore, postmenopausal women were selected as the study subjects to control the effects of lifestyle habits such as smoking and drinking, which are relatively more linked to aging and oxidative stress in women than in men. Therefore, it is significant that the study was conducted on postmenopausal women with decreased estrogen levels, which is related to the aging process and antioxidant capacity.

This study is the first randomized, double-blind, placebo-controlled study to investigate the comprehensive efficacy of KRG administration on biological aging, antioxidant activity, and clinical symptoms in Korean postmenopausal women. However, this study also has some limitations. The first limitation is that the KRG group was administered 2 g/day of KRG for 8 weeks. Previous clinical studies evaluated for

changes of mtDNA copy number, telomere length, antioxidant indicators, and clinical symptoms were varied with the dose of the KRG from 1 g/day to 6 g/day and the duration of administration from 2 weeks to 12 weeks. These ranges indicate that the dose and duration of KRG administration are not strongly agreed upon, so use of higher doses or longer administration durations in this study may have produced different outcomes, including LDL cholesterol, telomere length, oxidative stress, and MRS questionnaire responses. However, a daily dose of 2 g of KRG is generally available as a healthy functional supplement in the market. Accordingly, in this study, we selected 2 g per day, the dosage for safety KRG intake as a general health functional food. Therefore, determining the optimal dose and duration for the KRG intervention should be more investigated in the future. The second limitation is that, although we told the participants to keep their usual lifestyle during the clinical trial, we could not strictly control an individual`s behaviors, which could affect biological aging, oxidative stress, antioxidant capacity, and clinical symptoms. The third limitation is that the participants were only Korean postmenopausal women, so the results may not be broadly generalizable to other populations, such as men, premenopausal women, and different ethnicities.

V. CONCLUSION

This study examined the effects of KRG consumption on the primary outcomes variables of interest, which were changes in biological aging markers, oxidative stress, and antioxidant capacity, and secondary variables of interest, which were fatigue and menopausal symptoms, in Korean postmenopausal women. The results showed that administering 2 g/day of KRG for 8 weeks increased mtDNA copy number, increased

antioxidant activity, and reduced fatigue symptoms more than the placebo.

REFERENCES

1. Baeg IH, So SH. The world ginseng market and the ginseng (Korea). *J Ginseng Res* 2013;37:1-7.

2. So SH, Lee JW, Kim YS, Hyun SH, Han CK. Red ginseng monograph. *J Ginseng Res* 2018;42:549-561.

3. Levine ME, Lu AT, Chen BH, Hernandez DG, Singleton AB, Ferrucci L, et al. Menopause accelerates biological aging. *Proc Natl Acad Sci U S A* 2016;113:9327-9332.

4. Ossewaarde ME, Bots ML, Verbeek AL, Peeters PH, van der Graaf Y, Grobbee DE, et al. Age at menopause, cause-specific mortality and total life expectancy. *Epidemiology* 2005;16:556-562.

5. Kim HM, Park J, Ryu SY, Kim J. The effect of menopause on the metabolic syndrome among Korean women: the Korean National Health and Nutrition Examination Survey, 2001. *Diabetes Care* 2007;30:701-706.

6. Auro K, Joensuu A, Fischer K, Kettunen J, Salo P, Mattsson H, et al. A metabolic view on menopause and ageing. *Nat Commun* 2014;5:4708.

7. Eisenberg DT. An evolutionary review of human telomere biology: the thrifty telomere hypothesis and notes on potential adaptive paternal effects. *Am J Hum Biol* 2011;23:149-167.

8. Lopez-Otin C, Blasco MA, Partridge L, Serrano M, Kroemer G. The hallmarks of aging. *Cell* 2013;153:1194-1217.

9. Balaban RS, Nemoto S, Finkel T. Mitochondria, oxidants, and aging. *Cell* 2005;120:483-495.

10. Zhang R, Wang Y, Ye K, Picard M, Gu Z. Independent impacts of aging on

mitochondrial DNA quantity and quality in humans. *BMC Genomics* 2017;18:890.

11. Kim JY, Park JY, Kang HJ, Kim OY, Lee JH. Beneficial effects of Korean red ginseng on lymphocyte DNA damage, antioxidant enzyme activity, and LDL oxidation in healthy participants: a randomized, double-blind, placebo-controlled trial. *Nutr J* 2012;11:47.
12. Seo SK, Hong Y, Yun BH, Chon SJ, Jung YS, Park JH, et al. Antioxidative effects of Korean red ginseng in postmenopausal women: a double-blind randomized controlled trial. *J Ethnopharmacol* 2014;154:753-757.
13. Jung DH, Lee YJ, Kim CB, Kim JY, Shin SH, Park JK. Effects of ginseng on peripheral blood mitochondrial DNA copy number and hormones in men with metabolic syndrome: A randomized clinical and pilot study. *Complement Ther Med* 2016;24:40-46.
14. Liguori I, Russo G, Curcio F, Bulli G, Aran L, Della-Morte D, et al. Oxidative stress, aging, and diseases. *Clin Interv Aging* 2018;13:757-772.
15. Pandey KB, Rizvi SI. Markers of oxidative stress in erythrocytes and plasma during aging in humans. *Oxid Med Cell Longev* 2010;3:2-12.
16. Erel O. A novel automated direct measurement method for total antioxidant capacity using a new generation, more stable ABTS radical cation. *Clin Biochem* 2004;37:277-285.
17. Erel O. A new automated colorimetric method for measuring total oxidant status. *Clin Biochem* 2005;38:1103-1111.
18. Borras C, Gambini J, Lopez-Grueso R, Pallardo FV, Vina J. Direct antioxidant and protective effect of estradiol on isolated mitochondria. *Biochim Biophys Acta* 2010;1802:205-211.

19. Cawthon RM. Telomere length measurement by a novel monochrome multiplex quantitative PCR method. *Nucleic Acids Res* 2009;37:e21.

20. Krupp LB, LaRocca NG, Muir-Nash J, Steinberg AD. The fatigue severity scale. Application to patients with multiple sclerosis and systemic lupus erythematosus. *Arch Neurol* 1989;46:1121-1123.

21. Mills R, Young C, Nicholas R, Pallant J, Tennant A. Rasch analysis of the Fatigue Severity Scale in multiple sclerosis. *Mult Scler* 2009;15:81-87.

22. Mattsson M, Moller B, Lundberg I, Gard G, Bostrom C. Reliability and validity of the Fatigue Severity Scale in Swedish for patients with systemic lupus erythematosus. *Scand J Rheumatol* 2008;37:269-277.

23. Herlofson K, Larsen JP. Measuring fatigue in patients with Parkinson's disease - the Fatigue Severity Scale. *Eur J Neurol* 2002;9:595-600.

24. Hauser GA, Huber IC, Keller PJ, Lauritzen C, Schneider HP. [Evaluation of climacteric symptoms (Menopause Rating Scale)]. *Zentralbl Gynakol* 1994;116:16-23.

25. Heinemann K, Ruebig A, Potthoff P, Schneider HP, Strelow F, Heinemann LA, et al. The Menopause Rating Scale (MRS) scale: a methodological review. *Health Qual Life Outcomes* 2004;2:45.

26. Shin EJ, Jo S, Choi S, Cho CW, Lim WC, Hong HD, et al. Red Ginseng Improves Exercise Endurance by Promoting Mitochondrial Biogenesis and Myoblast Differentiation. *Molecules* 2020;25.

27. Park JK, Shim JY, Cho AR, Cho MR, Lee YJ. Korean Red Ginseng Protects Against Mitochondrial Damage and Intracellular Inflammation in an Animal Model of Type 2 Diabetes Mellitus. *J Med Food* 2018;21:544-550.

28. Taherzadeh-Fard E, Saft C, Akkad DA, Wieczorek S, Haghikia A, Chan A, et al.

PGC-1alpha downstream transcription factors NRF-1 and TFAM are genetic modifiers of Huntington disease. *Mol Neurodegener* 2011;6:32.

29. Gureev AP, Shaforostova EA, Popov VN. Regulation of Mitochondrial Biogenesis as a Way for Active Longevity: Interaction Between the Nrf2 and PGC-1alpha Signaling Pathways. *Front Genet* 2019;10:435.
30. Kim MJ, Koo YD, Kim M, Lim S, Park YJ, Chung SS, et al. Rg3 Improves Mitochondrial Function and the Expression of Key Genes Involved in Mitochondrial Biogenesis in C2C12 Myotubes. *Diabetes Metab J* 2016;40:406-413.
31. Wallace DC. A mitochondrial paradigm of metabolic and degenerative diseases, aging, and cancer: a dawn for evolutionary medicine. *Annu Rev Genet* 2005;39:359-407.
32. Kim YS, Kim YH, Noh JR, Cho ES, Park JH, Son HY. Protective Effect of Korean Red Ginseng against Aflatoxin B1-Induced Hepatotoxicity in Rat. *J Ginseng Res* 2011;35:243-249.
33. Ramesh T, Kim SW, Hwang SY, Sohn SH, Yoo SK, Kim SK. Panax ginseng reduces oxidative stress and restores antioxidant capacity in aged rats. *Nutr Res* 2012;32:718-726.
34. Yu SY, Yoon BR, Lee YJ, Lee JS, Hong HD, Lee YC, et al. Inhibitory effect of high temperature- and high pressure-treated red ginseng on exercise-induced oxidative stress in ICR mouse. *Nutrients* 2014;6:1003-1015.
35. Bak MJ, Jun M, Jeong WS. Antioxidant and hepatoprotective effects of the red ginseng essential oil in H(2)O(2)-treated hepG2 cells and CCl(4)-treated mice. *Int J Mol Sci* 2012;13:2314-2330.
36. Park HM, Kim SJ, Mun AR, Go HK, Kim GB, Kim SZ, et al. Korean red ginseng

and its primary ginsenosides inhibit ethanol-induced oxidative injury by suppression of the MAPK pathway in TIB-73 cells. *J Ethnopharmacol* 2012;141:1071-1076.

37. Park SH, Jang JH, Chen CY, Na HK, Surh YJ. A formulated red ginseng extract rescues PC12 cells from PCB-induced oxidative cell death through Nrf2-mediated upregulation of heme oxygenase-1 and glutamate cysteine ligase. *Toxicology* 2010;278:131-139.
38. Dong GZ, Jang EJ, Kang SH, Cho IJ, Park SD, Kim SC, et al. Red ginseng abrogates oxidative stress via mitochondria protection mediated by LKB1-AMPK pathway. *BMC Complement Altern Med* 2013;13:64.
39. Bach HV, Kim J, Myung SK, Cho YA. Efficacy of Ginseng Supplements on Fatigue and Physical Performance: a Meta-analysis. *J Korean Med Sci* 2016;31:1879-1886.
40. Sung WS, Kang HR, Jung CY, Park SS, Lee SH, Kim EJ. Efficacy of Korean red ginseng (Panax ginseng) for middle-aged and moderate level of chronic fatigue patients: A randomized, double-blind, placebo-controlled trial. *Complement Ther Med* 2020;48:102246.
41. Park BJ, Lim YS, Lee HJ, Eum WS, Park J, Han KH, et al. Anti-oxidative effects of Phellinus linteus and red ginseng extracts on oxidative stress-induced DNA damage. *BMB Rep* 2009;42:500-505.
42. Lee JH, Sul D, Oh E, Jung WW, Hwang KW, Hwang TS, et al. Panax ginseng effects on DNA damage, CYP1A1 expression and histopathological changes in testes of rats exposed to 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Food Chem Toxicol* 2007;45:2237-2244.
43. Kim SH, Park KS. Effects of Panax ginseng extract on lipid metabolism in humans.

Pharmacol Res 2003;48:511-513.

44. Delui MH, Fatehi H, Manavifar M, Amini M, Ghayour-Mobarhan M, Zahedi M, et al. The Effects of Panax Ginseng on Lipid Profile, Pro-oxidant: Antioxidant Status and High-sensitivity C Reactive Protein Levels in Hyperlipidemic Patients in Iran. *Int J Prev Med* 2013;4:1045-1051.

Appendix 1.

Fatigue severity scale

다음 각 문항에 본인의 상태를 가장 잘 나타내는 점수에 표시해 주십시요. (1점= 문항의 내용에 대하여 강력하게 반대한다) (7점= 문항의 내용에 대하여 강력하게 동의한다)							
지난 한 주 동안 나는 아래와 같은 항목을 발견하였다.	1점	2점	3점	4점	5점	6점	7점
1. 내가 피로감을 느낄 때 나의 의욕은 매우 떨어져 있다.							
2. 운동을 하면 피로해진다.							
3. 나는 쉽게 피곤해진다.							
4. 피로로 인해 나의 신체적 기능이 떨어진다고 느낀다							
5. 피곤함은 나에게 여러 가지 문제들을 빈번하게 일으킨다.							
6. 피곤은 나의 신체적 기능을 지속적으로 유지하기 어렵게 한다							
7. 피곤함은 나의 의무와 책무를 수행하는데 있어 방해를 끼치고 있다.							
8. 피곤함은 나에게 무력감을 주는 3가지 가장 흔한 증상 중 하나이다							
9. 피곤함은 나의 일과 가족, 사회생활에 악영향을 준다.							

Appendix 2.

Menopause rating scale

아래의 증상 중 현재 본인에게 해당하는 증상은 무엇인지 체크하세요. 각 항목당 본인의 증상 정도에 따라서 체크하시면 됩니다. 해당 항목에 증상이 없는 경우, '없음' 에 표시하시기 바랍니다.					
	없음 (0)	약간 (1)	보통 (2)	심함 (3)	매우 심함 (4)
1. 안면홍조 및 발한이 있다. (갑작스럽게 덥거나 땀이 나는 경험)					
2. 심장 불편감이 있다. (갑자기 가슴이 빨리 뛰거나,불규칙하게 뛰거나 답답한 느낌)					
3. 수면문제 (잠들기 어렵거나 수면의 질이 좋지 않음)					
4. 우울한 기분 (슬프거나 의욕이 없고 쉽게 눈물이 나고 감정이 수시로 변함)					
5. 과민성 (신경이 예민해지거나 긴장감, 공격적으로 되는 느낌)					
6. 불안감 (초조함이나 공황상태가 되는 느낌)					
7. 신체 및 정신적 피로 (일상생활 수행 능력이 떨어지거나 기억력, 집중력 감소)					

8. 성적인 문제 (성욕이 떨어지고 성적 만족감의 변화)					
9. 배뇨 문제 (소변 불편감, 자주 마려움, 요실금)					
10. 질 건조감 (건조감. 화끈거림, 성교시 불편감)					
11. 관절 및 근육 불편감 (관절 통증 및 근육 통증)					

ABSTRACT (IN KOREAN)

홍삼이 한국인 폐경 후 여성에서 생물학적 노화 지표와 항산화에 미치는 효과: 이중 맹검 무작위 대조 연구

<지도교수 이용제>

연세대학교 대학원 의학과

정태하

배경: 폐경 후 여성은 에스트로겐 호르몬의 감소로 인해 노화 및 산화 스트레스에 취약하다고 알려져 있다. 이전 연구들에서 고려 홍삼이 노화와 항산화 능력에 유익한 효과가 있다고 보고하였다. 따라서 우리는 폐경 후 여성에서 생물학적 노화, 산화 스트레스 및 항산화 능력에 대한 고려 홍삼의 효과를 보고자 하였다.

방법: 본 연구는 8주 동안 무작위 이중 맹검 위약 대조군 연구로 진행이 되었다. 총 73명이 연구 대상자가 되었고 고려 홍삼군과 위약군을 1:1 무작위 배정하여 연구를 진행하였다. 생물학적 노화 지표로 telomere length 와 mitochondria (mt) DNA copy number, 산화 스트레스 및 항산화 능력의 지표로 total oxidant status (TOS) 및 total antioxidant status (TAS)를 평가하였다. 또한 중재 전과 후의 피로와 갱년기의 임상증상을 평가하기 위해 fatigue severity scale (FSS)과 menopause rating scale (MRS)을 비교하였다.

결과: 총 63명의 참가자(고려 홍삼군=33명, 위약군=30명)가 연구를 완료했다. mtDNA copy number의 중재 전후 변화량은 고려 홍삼군에서 1.58±2.05, 위약군에서 0.28±2.36로 고려 홍삼군에서 더 증가하였고 두 그룹간의 통계적 차이를 보였다(p=0.023). 또, 항산화 능력인 TAS의 변화량은 고려 홍삼군에서 0.11±0.25mmol/L 로 증가하였고 위약군에서는 -0.04±0.16mmol/L로 감소하였으며, 두 그룹간의 유의한 차이를 보였다(p=0.011). 피로도에 대한 임상 증상 지표인 FSS는 고려 홍삼군과 위약군 모두 감소하였지만(-7±12 vs -1±11), 홍삼군에서 p=0.002로 의미있는 감소를 보였고 두 그룹간의 통계적으로 유의미한 차이를 보였다(p=0.033).

결론: 8주 동안 고려 홍삼(2g/하루)의 복용은 폐경 후 여성에서 생물학적 노화 지표인 mtDNA copy number, 항산화 능력인 TAS 및 피로 증상을 유의하게 증가시켰다.

핵심되는 말: 고려 홍삼; 산화 스트레스; 항산화 능력; 생물학적 나이; 폐경 후 여성