보건학석사 학위논문

# The effects of Korean-DASH diet on metabolic syndrome, insulin resistance and gut microbiota

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2016 년 8 월

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지도교수 성주헌

이 논문을 보건학 석사학위논문으로 제출함
2016년 7월

서울대학교 대학원
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채련의 석사학위논문을 인준함
2016년 7월

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# Abstract

**Introduction:** The prevalence of metabolic syndrome (MetS) has increased in Korea and it has been important issue in public health. The risk of MetS is associated with lifestyle and Dietary pattern is important factor to prevent and control the MetS. In addition, the intestinal microbiota is modulated by host diet and affects human metabolic health. The aim of this study is to assess the influence of a Korean DASH diet on metabolic syndrome, insulin resistance improvement in Korean adults and the changes of intestinal microbiota composition and diversity.

**Methods:** Based on Dietary Approaches to Stop Hypertension (DASH) diet patterns, the modified traditional Korean diet was developed. This K-DASH diet is rich in grains, vegetables, nuts, poultry and low in sodium and red meat. K-DASH diet intervention was conducted during 10-days with two sub groups of traditional Korean diet based Korean (TK group) who have been adhering traditional Korean diet and Western diet based Korean (WK group) who have been adhering western diet. A total 65 subjects participated in 10-days intervention. (TK group n=46, WK group n=19) We collected data of anthropometric, biochemical measurements and microbial materials at pre-post intervention each. The microbial sequencing were completed by the Illumina Miseq platform and after quality check the sequences, quantitative metagenomics analysis were performed using QIIME 1.8.0 software.

**Results:** During 10-days K-DASH diet intervention the intakes of protein and fiber were significantly increased and the dietary sodium intakes were

decreased ($p<0.05$). In metabolic syndrome indicators, the body weight and waist circumference were decreased (-0.90kg, -0.42cm; $p<0.001$) and systolic blood pressure (SBP) and diastolic blood pressure (DBP) were reduced in whole subjects (-4.85mmHg, -2.45mmHg; $p<0.01$). Also the total cholesterol and triglycerides level were significantly decreased (-14.4mmo/L, -14.3 mmol/L; $p<0.001$). Furthermore, there were significant reduction in insulin resistance index HOMA-IR and fasting insulin level. (-0.48, -2.30 pmol/L; $p<0.01$) Comparison of gut microbiota composition between groups showed that TK group have more diverse microbiota than WK group and TK group have relatively higher in *Bacteroidetes* and lower in *Firmicutes* than WK group. After 10-days K-DASH intervention the microbiota diversity and composition were altered including decreased *Firmicutes / Bacteroidetes* ratio and increased alpha diversity. And the changes of F/B ratio and the abundances of *L.reuteri, L.zeae, C. butyricum, Roseburia sp* were positively correlated with insulin level and HOMA-IR ($r>0.65$, $p<0.01$)

**Discussion:** These results suggest that K-DASH diet could play a protective role in the treatment of metabolic syndrome in Korean. The K-DASH diet was modified to remedy the disadvantages of traditional Korean diet, on this account K-DASH had an effect on both group. Dietary modulation altered gut microbiota composition and the carbohydrate dependent microbial abundance was reduced. Even though the absolute amount of nutrient was not changes, the relative amount of nutrient could affect the microbiota composition. For the more efficient treatment of metabolic syndrome, comprehensive lifestyle modification is needed with K-DASH diet.

**Keywords:** Metabolic syndrome, Intestinal microbiota, Korean Dietary Approach to Stop Hypertension (K-DASH), Metagenomics, Dietary intervention, The healthy Korean diet

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# I. INTRODUCTION

## 1. Background

Metabolic Syndrome(MetS) is a clustering of several cardio-metabolic risk factors that is related to obesity(especially central adiposity), dysglycemia, elevated blood pressure, low high-density lipoprotein cholesterol(HDL-C)level and elevated triglyceride levels.[1] MetS is associated with development of various chronic diseases such as cardiovascular disease and diabetes mellitus.[2, 3]

The prevalence of MetS has increased worldwide and also Korean have an increasing tendency toward MetS.[2] According to the Korea National Health and Nutrition Examination Survey (KNHANES), the prevalence of MetS have significantly increased from 24.9% in 1998, 29.2% in 2001 and 30.4% in 2005 to 31.3% in 2007 ($P<0.01$)[2] The rapid socioeconomic growth of Korea have led changes of lifestyle such as westernized diet and sedentary behavior, which led to an increasing MetS. [3, 4] The risk of MetS is associated with and unhealthy lifestyle and particularly diets.[4-8] Thus, lifestyle and dietary patterns modification is generally regarded as a key intervention in MetS.[6] Especially, the effect of diet modification that are rich in fruits, vegetables, whole grains and low-fat dairy products on MetS has been highlighted in many clinical and epidemiological studies.[9] The Korean traditional diet is consist of high in carbohydrate, low in fat and abundant in vegetables, fruits and fermented products, therefore it is likely to have a protective effect on metabolic syndrome. A variety of studies have been conducted to discover the association

between Korean traditional diet and MetS, however these studies were carried out by observational study and the traditional patterns showed no association with metabolic syndrome. [6, 8, 10, 11]. Therefore, it is important to understand the mechanism of metabolic syndrome development process more comprehensively, in order to make Korean specific MetS prevention strategy.

Meanwhile, many studies have proposed the potential role of intestinal microbiota on human metabolic disorders and inflammation. The human body contains 100 trillion bacteria, 10-fold greater than whole human body cells, that are modulated by diet. [12] Gut microbiota have a various function: regulate host nutrition and energy harvest by the production of vitamins and fermentation and influence of intestinal homeostasis and development of host immune system. [13] Compositional changes of human intestinal microbiota in response to weight changes and dietary changes have been examined. [14, 15] The obese group have a relative low abundance in *Bacteroidetes* and high in *Firmicutes* and also the weight loss and low-calorie diet was associated with this ratio. [14] And gut microbiota dysbiosis may lead to various human disease including Irritable Bowel Disease, Crohn's disease, type 2 diabetes and gastric cancers. [16] Accordingly, the gut microbiota interact with diet components and host health and play a key role in human metabolic pathology. [7, 9, 14, 17]

The evidence is more needed to establish a causal relationship between dietary intervention and alternation of gut microbiota and human metabolic syndrome in Korean population. Therefore, the aim of this study is to develop modified healthy Korean traditional diet and to examine the effect on metabolic syndrome as well as the changes of intestinal microbiota composition.

This research was supported by the Globalization of Korean Foods R&D program, funded by the Ministry of Food, Agriculture, Forestry and Fisheries (IPET).

### 2. Objective

This study aims to identify changes in metabolic indicators and health effects of modified Korean DASH diets. And to establish a relationship among diet, microbiota and human health. Specific aims of the study are (1) to develop the modified Korean Dietary Approach to Stop Hypertension (DASH), (2) to evaluate the effect of K-DASH on metabolic syndrome and microbiota composition (3) to identify the role of microbiota in human health and the relationship.

## II. METHODS

### 1. Study population

The participants were recruited from the Healthy Twin Study, a prospective cohort study population that is composed of Korean adult twin and their family members. Forty-two independent families were included in this reaearch. The subjects were restricted to those who aged 30 to 50 years, have never been diagnosed with diabetes, cancer, cardiovascular disease and those who can intake lunch box for ten days.(**Table1**)

A total of 66 participants were recruited from Health Twin Study and Korean adoptee community. Participants agreed to dietary intervention and a serials of examinations including clinical test, anthropometry investigation, demographic questionnaire and informed consent was obtained. Participants who was absent at secondary examination was excluded from this study (n=1). A total 65 adults (Korean twin adults=46 and Korean adoptee=19, 8 men and 57 women) were included in the final data analysis.

**Table 1.** Inclusion criteria for subject recruitment.

| Inclusion criteria |
| --- |
| · Males and Females aged between 30 and 50 years |
| · Body Mass Index (BMI) > 18.5 $kg/m^{-2}$ |
| · Never been diagnosed with cardiovascular disease, diabetes, cancer. |
| · Had received therapeutic antibiotics to treat bacterial infections |

## 2. Dietary intervention

A modified K-DASH (Korean Dietary Approach to Stop Hypertension) were developed by nutrition research team of Sookmyung women's university led by Professor So-yeon Jin and Jeong-eun Lee.

*Assessment of dietary intake*

Participants were asked to record their food intake during 3 days in ‘three day food record’ before the intervention. A nutritionist gave instructions for recording the food intakes and interviewed the subjects about the completion

of the record. Based on the record, nutritionist team adjusted their diet plan. Dietary intervention was conducted for 10 days and two lunch boxes per day were delivered. During the intervention, subjects took and sent their lunch box pictures to check their food intakes. And all food except the lunch box were checked by food recording or mobile phone pictures.

*Korean DASH diet plans*

Korean DASH diet is modified Korean healthy diet for reducing risk of metabolic disorders. The original DASH diet plan was developed to lower blood pressure by the US National Institutes of Health.[18] Like original DASH diet plan this K-DASH plan was developed to prevent hypertension and metabolic syndrome by modifying the traditional Korean diets. Korean DASH Diet is rich in grains, fruits and vegetables; lower sodium content; lower red meats, rich in fish and poultry; nuts and bean. It is consist of high fiber and low in saturated fat. K-DASH was tested during a 10-days with two sub-groups of traditional Korean diet Korean (TK) and western diet Korean (WK). The research team delivered the customized lunch boxes to participants for 10-days. With lunch boxes, the subjects were provided dietary guideline and recipes of K-DASH diet plan.

Recipes and intakes for the K-DASH diets were analyzed using Can Pro 4.0 nutrients databases. (The Korean Nutrition Society, Korea)

## 3. Data Collection

All the participants were undertaken clinical examinations between baseline and at 10 days. The clinical examinations comprised demographic investigation, anthropometric investigation, blood and urine sample tests, dietary assessments. The data were obtained using standard protocols of clinical laboratory of Samsung medical center. This study was approved by Korean Institutional Review Board and written consent was obtained from all subjects.

*Demographic investigations*

Socio-demographic variables were obtained by general questionnaire. The questionnaire included such as name, age, sex, familial relationship, familial disease history and education, job and health related variables such as medical history, physical activity, smoking status, alcohol intake.

*Anthropometry investigations*

Weight and height were measured using a digital scale and Body mass index (BMI) was calculated from the measured weight (kg)/height ($m^2$). Hip circumference was determined at the widest point and Waist circumference was measured at the midpoint between last rip and the upper upper edge of pelvis. Blood pressure SBP and DBP were measured by sphygmomanometer three tiems with a 5 min interval and 10-min seated rest. All the anthropometry measurements were conducted two or three times to arrive at a mean values.

*Metabolic indicators*

Blood samples were collected after 12 hours fasting and urine sample were collected a 12-hour urine specimen. Blood samples were collected in an EDTA-coated tube and SST tube and centrifuged at 3000rpm for 10 mins. Plasma and Serum collected in EDTA tube and SST tube each, were divided into microfuge and stored at 2-8℃ until analysis. A complete blood count test (CBC; WBC, RBC, Hb, Hct, MCV, MCH, MCHC, Platelet) was analyzed by flow cytometry. Blood biochemical test included level of total cholesterol, high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol (LDL-C), triglycerides(TG), HbA1C, apolipoprotein A and B, adiponectin. All lipid levels in blood were analyzed by enzyme method. Serum glucose level was estimated by colorimeter and plasma insulin was measured by insulin radioimmunoassay.

*Insulin sensitivity and resistance index*

To assess insulin sensitivity and resistance, HOMA-IR, HOMA-βcell, QUICKI, ISI index were used. HOMA-IR was calculated according to the formula: [fasting insulin (μIU/ml)**x**fasting glucose (nmol/L)/22.5] [19] HOMA-βcell, estimating beta cell secretory function, was calculated [20**x**fasting insulin (μIU/ml) / fasting glucose (nmol/L)-3.5] [20] QUICKI index, quantitative insulin sensitivity check index, was calculated as 1/[log fasting insulin (μU/mL) +log fasting glucose (mg/dL)] [21] ISI, insulin sensitivity index, was calculated as exp [3.29-0.25ln(fasting insulin) – 0.22ln(BMI)-0.28ln(TG)] [22]

## 4. Metabolic syndrome definition

Metabolic syndrome was defined according to the National Cholesterol Education Program Adult Treatment PanelⅢ.[23] As Asian have smaller body size than Caucasians, waist circumference was determined using Asian guidelines.[24] Participants who had Any three of more of the following criteria were diagnosed as MetS: 1) high waist circumference, waist circumference >90cm in men and >85cm in women; elevated blood pressure, systolic blood pressure(SBP) ≥130mmHg and diastolic blood pressure(DBP) ≥85mmHg; Low high-density lipoprotein cholesterol (HDL-C) level, <40mg/dL in men and <50mg/dL in women; elevated triglycerides(TG) level, ≥150mg/dL (1.69 mmol/L); elevated fasting blood glucose level, >100 mg/dL.

## 5. Microbiome analysis

In order to discover whether K-DASH diet impacts on the intestinal microbiota composition and diversity, we analyzed the changes of microbiota during 10-days intervention.

*DNA Extraction, Amplication and Sequencing*

Fecal samples collected from the participants were placed at 4℃ and transferred freezer -80℃ within 24 hours after produced. Fecal microbiota DNA was extracted using bead beating protocol and QIAamp DNA Stool Mini Kit (Qiagen, Germany). The extracted from each sample was used as template to amplify the V1-V3 region of the 16S rRNA by PCR. Amplicons were

purified using Ultra Clean PCR Clean-Up Kit (MO BIO Laboratories, USA) and quantified PCR library using KAPA Library Quant Kit (Kapa Biosystems, USA).

*Analysis of Microbiota*

Amplified genes were sequenced on an Illumina Miseq. For quality control, all the singletons and chimeras in raw data were removed using Uchime.[25, 26] A qualified pyrosequencing data were picked to 97% OTUs against the Greengenes reference database[27] and the remains were clustered *de novo* using Usearch.[28] Taxonomy of each OTUs were assigned using RDP classfier [29] and built PCoA beta diversity and Alpha diversity using UniFrac.[30] All microbiota sequence processing was performed by QIIME Ver 1.8.0.[31]

## 6. Statistical analysis

Demographic and health related variable description were presented as mean ± standard deviation for continuous variables and counts, percentages for categorical variables. To compare difference by sex the Student t-test for continuous variables and $\chi^2$ test for categorical variables were used.

A paired Student's *t*-test was used to compare differences baseline and post intervention on the nutrients variables, anthropometric, metabolic indicators and insulin indexes. When variables were not normally distributed, non-parametric methods were used. The Mann-Whitney U test was used to compare between group difference and Wilcoxon signed-ranked test was used to

compare the pre-post intervention changes.

To determine the effect of the K-DASH diet on metabolic syndrome, biochemical variables and insulin indexes within and between groups, linear mixed effect model was used. The model included subjects as a random effects and age, sex, study group, time point(pre-post) and time point*study group interaction as covariates. Analyses of nutrients intakes included body weight as covariate too. *P*-value for fixed effects was computed using Satterthwaite's approximations. The changes in phylum, genus, species level in intestinal microbiota between baseline and 10-days after the intervention were correlated to changes in metabolic indicators, biochemical variables and insulin indexes by using Pearson correlation method.

All statistical analyses were performed using the SAS survey procedure (Version 9.3 SAS institute, Cary, NC).

# III. RESULTS

## 1. Descriptive Statistics

Sixty-five participants (TK group n=46, WK group n=19) were included for analysis. The subjects consisted of 6 males and 57 females with mean ages of 31.0 ± 5.81 and 45.9 ± 10.7 each. **Table2** present the general characteristics of the study subjects by sex.

Generally, Men had higher baseline values for height, body weight, BMI, waist circumference, systolic and diastolic blood pressures and lower baseline values in ages. The proportion of current smoker was higher in men than women. There was no sex difference in alcohol intakes and physical activity.

Among the 65 participants 13 had metabolic syndrome. The prevalence of MetS was 20.0% (37.5% in men and 17.5% in women). The prevalence of the MetS and the number of the metabolic syndrome criteria were not significantly different between sexes. No sex difference were observed regarding waist circumference, HDL-cholesterol, Triglycerides, fasting blood glucose criteria, Only elevated blood pressure was significantly greater in men than women.

**Table 2.** Descriptive characteristics of the study population.

| Characteristics | Men (n=8) | Women (n=57) | *p*-value |
|---|---|---|---|
| **Age (year)** | 31.0 ± 5.81 | 45.9 ± 10.7 | <0.001 |
| **Height (cm)** | 165.5 ± 15.1 | 157.1 ± 5.4 | <0.001 |
| **Weight (kg)** | 72.1 ± 15.1 | 57.9 ± 10.8 | 0.002 |
| **BMI (kg/m²)** | 26.2 ± 4.87 | 23.4 ± 3.62 | <0.001 |
| **Waist circumference (cm)** | 84.7 ± 12.9 | 80.4 ± 8.57 | <0.001 |
| **Hip circumference (cm)** | 100.2 ± 7.38 | 95.7 ± 7.67 | 0.055 |
| **Systolic blood pressure (mmHg)** | 125.9 ± 8.63 | 114.8 ± 15.0 | 0.047 |
| **Diastolic blood pressure (mmHg)** | 85.9 ± 10.4 | 74.2 ± 10.2 | 0.003 |
| **Current smokers** | 3 (37.5%) | 3 (5.26%) | 0.003 |
| **Current alcohol intake** | 5 (62.5%) | 19 (33.3%) | 0.109 |
| **Physical activity** | | | 0.558 |
| Never or rarely | 1 (12.5%) | 12 (21.0%) | |
| Less than 1/week | 2 (25.0%) | 16 (28.1%) | |
| 1-2 /week | 4 (50.0%) | 15 (26.3%) | |
| More than 3 times a week | 1 (12.5%) | 14 (24.6%) | |

| | | | |
|---|---|---|---|
| **Prevalence of Metabolic syndrome** | | | |
| High waist circumference | 3 (37.5%) | 22 (38.6%) | 0.952 |
| Elevated blood pressure | 5 (71.4%) | 12 (21.1%) | 0.004 |
| Low HDL-Cholesterol | 4 (50.0%) | 23 (40.3%) | 0.604 |
| Elevated triglyceride | 0 (0.00%) | 9 (15.8%) | 0.226 |
| Elevated fasting blood glucose | 1 (12.5%) | 9 (15.8%) | 0.809 |
| **Metabolic syndrome** | 3 (37.5%) | 10 (17.5%) | 0.186 |

Continuous data were described in mean ± standard deviation P values were obtained from t-test. Categorical data were presented in counts and tested with Pearson's Chi-square test.

## 2. Difference in Nutrients intakes.

*Whole subject difference in nutrient intakes during the intervention*

Dietary total energy intake was varied according to each participant's RDI (recommended daily intakes). During the 10-days K-DASH diet intervention, there was no significant differences in estimated energy calories compared to baseline. The intakes of Carbohydrate and total lipid were decreased, but it was not a significant change.

**Table 3.** Changes in dietary intakes during the 10-days dietary intervention

| | Whole group (n=65) | | | |
|---|---|---|---|---|
| | Baseline mean± SD | 10-days mean± SD | Mean difference | P-value[1] |
| Calorie (kcal) | 1538.3±400.5 | 1462.4±226.3 | -75.89±469.7 | 0.220 |
| Carbohydrate (g/d) | 240.2±61.8 | 228.5±41.3 | -11.64±75.48 | 0.241 |
| Total lipid (g/d) | 40.2±18.5 | 35±6.4 | -5.15±19.84 | 0.051 |
| Total protein (g/d) | 60.2±19.0 | 73.6±9.3 | 13.39±21.39 | <0.001** |
| Cholesterol (mg/d) | 259.4±156.3 | 250±75.2 | -9.41±171.5 | 0.675 |
| Fiber (g/d) | 20.1±6.5 | 28±4.8 | 7.86±7.34 | <0.001** |
| Sodium (mg) | 3069.5±1177.4 | 1989.7±500.6 | -1079.8±1247 | <0.001** |
| Vitamin A (ug RE/d) | 902.5±545.6 | 1154±265.7 | 251.5±506.7 | <0.001** |
| Thiamin (mg/d) | 1.1±0.3 | 1.2±0.2 | 0.1±0.37 | 0.047* |
| Riboflavin (mg/d) | 1.0±0.3 | 1.2±0.2 | 0.26±0.38 | <0.001** |
| Vitamin C (mg/d) | 108.9±81.4 | 170.1±44.7 | 61.21±90.8 | <0.001** |
| Vitamin E (mg/d) | 14.0±7.2 | 14.2±2.4 | 0.15±7.43 | 0.877 |
| Niacin (mg/d) | 14.5±4.5 | 16.8±2.5 | 2.31±5.28 | 0.001** |
| Calcium (mg/d) | 373.6±137.9 | 464±80.7 | 90.42±166.9 | <0.001** |
| Potassium (mg/d) | 2507.6±742.6 | 3649.5±601.5 | 1141.9±866.4 | <0.001** |
| Phosphorus (mg/d) | 923.9±250.6 | 1186.3±168.7 | 262.3±297.7 | <0.001** |
| Iron (mg/d) | 13.6±5.0 | 16.1±2.3 | 2.58±5.56 | 0.001** |

All values are presented as means ± SD. * indicates statistically significant values (* p<.05 , ** p <.01)
[1]Paired Student's *t*-test was used for comparison baseline versus 10-days in dietary intakes.

Whereas, the consumption of dietary protein and Fiber was significantly increased. ($p<0.05$) The intakes of dietary sodium was significantly decreased. (-1079.8mg/d, $p<0.05$) and Vitamin A, Thiamin, Riboflavin, Vitamin C, Niacin, Calcium, Potassium, Phosphorus, lron were was significantly increased ($p<0.05$). **Table3** present the changes in dietary intakes during the 10-days dietary intervention.

*Baseline dietary patterns of the group.*

Two group of subjects have different diet style. The Traditional Korean style diet group (TK group) have been eating the traditional Korean diet such as rice, kimchi, soup, fermented soy-based condiments, raw or cooked vegetables.[6] Whereas the western style diet group (WK group) have lived abroad for a while and have been eating the western diet such as high intakes of red meat, processed meat, french fries, refined grains, high sugar drink and desserts.[32]

The baseline dietary patterns in both group is presented in Table4. The total caloric intakes was higher and carbohydrate intakes lower in the WK group compared with TK group but it was not a significant changes. In addition, the intakes of total lipid, protein, cholesterol was significantly higher in the WK group and the intakes of fiber was significantly higher in TK group. ($p<0.05$) No significant difference was observed in intakes of sodium, vitamins, calcium, potassium and Iron.

**Table 4.** Comparison of baseline dietary patterns between groups

| | TK group | WK group | Group difference | |
|---|---|---|---|---|
| | Baseline mean± SD | Baseline mean± SD | Mean± SD difference | P-value |
| Calorie (kcal) | 1487.2±279.7 | 1718.9±660.7 | 231.7±392.0 | 0.239 |
| Carbohydrate (g/d) | 242.6±51.1 | 231.5±92.4 | -11.1±62.1 | 0.683 |
| Total lipid (g/d) | 35.9±12.6 | 55.4±27.1 | 19.5±16.7 | 0.025* |
| Total protein (g/d) | 56.4±14.2 | 73.6±27.1 | 17.2±17.7 | 0.045* |
| Cholesterol (mg/d) | 225.2±124.3 | 380.7±199.1 | 155.5±143.3 | 0.018* |
| Fiber (g/d) | 21.5±6.3 | 15.3±4.8 | -6.2±6.0 | 0.002** |
| Sodium (mg) | 3029.2±1101.1 | 3212±1457.9 | 182.8±1185 | 0.625 |
| Vitamin A (ug RE/d) | 945.5±583.7 | 750.4±359.4 | -195.1±544 | 0.258 |
| Thiamin (mg/d) | 1.1±0.3 | 1.2±0.4 | 0.1±0.3 | 0.259 |
| Riboflavin (mg/d) | 1±0.3 | 1.1±0.4 | 0.2±0.3 | 0.074 |
| Vitamin C (mg/d) | 115.1±81.4 | 86.8±80.6 | -28.3±81.3 | 0.272 |
| Vitamin E (mg/d) | 13.2±4 | 17±13.2 | 3.8±7.0 | 0.325 |
| Niacin (mg/d) | 13.6±3.8 | 17.8±5.4 | 4.3±4.2 | 0.002** |
| Calcium (mg/d) | 376.7±128.1 | 362.6±173.8 | -14.1±138.9 | 0.747 |
| Potassium (mg/d) | 2540.3±786.7 | 2392.1±570.7 | -148.2±746 | 0.53 |
| Phosphorus (mg/d) | 919±225.2 | 941.3±336.0 | 22.2±252.6 | 0.78 |
| Iron (mg/d) | 13±3.9 | 15.5±7.6 | 2.4±4.9 | 0.282 |

All values are presented as means ± SD. TK group = Traditional diet style based Korean group. WK group = Western diet style based Korean group.
P value were determined by independent *t*-test between baseline values of TK group and WK group.
* indicates statistically significant values (* p<.05 , ** p <.01)

*Between group difference in nutrient intakes*

Table5. shows the changes in dietary intakes by group during 10-days K-DASH intervention. In TK group, the intakes of protein and fiber was significantly increased (18.49 g/d and 7.10 g/d respectively, $p<0.05$) and sodium intake was decreased (-1011.5 mg/d, $p<0.001$). And there were no significant difference in intakes of total calories, carbohydrate, total lipid and cholesterol. In WK group, the intake of fiber was significantly increased (10.56 g/d, $p<0.001$) and intakes of total lipid, cholesterol and sodium were decreased (-24.1 g/d, -143 mg/d and -1321 mg/d respectively, $p <0.05$).

In micronutrients, the intakes of vitamin C, vitamin A, Potassium were significantly increased in both intervention group. ($p<0.05$) In addition, the intakes of thiamin, riboflavin, niacin, calcium, phosphorus and Iron were significantly increased in TK group ($p<0.05$).

A significant interaction between time and group was observed in the intake of total calorie, total lipid, protein, cholesterol and niacin ($p<0.01$). The difference in post-pre difference according to group were observed in the intakes of total lipid, total protein, cholesterol and niacin ($p<0.05$)

**Table 5.** Changes in dietary intakes between baseline and 10-days intervention by group

| | TK group (n=46) | | | WK group (n=19) | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Baseline mean± SD | 10-days mean± SD | Mean difference | Baseline mean± SD | 10-days mean± SD | Mean difference | P value[a] | P value[b] | P value[c] | P value[d] |
| Calorie (kcal) | 1487.2±279.7 | 1494.3±212.2 | 7.09±329.3 | 1719±660.7 | 1349±246.7 | -369.5±734.8 | 0.095 | 0.041* | 0.913 | 0.008** |
| Carbohydrate (g/d) | 242.6±51.1 | 233.4±39.3 | -9.21±63.2 | 231.5±92.4 | 211.2±45.4 | -20.25±111.6 | 0.738 | 0.182 | 0.396 | 0.632 |
| Total lipid (g/d) | 35.9±12.6 | 36.1±5.9 | 0.19±13.1 | 55.4±27.1 | 31.3±6.8 | -24.06±27.6 | 0.009** | 0.015* | 0.941 | <.001** |
| Total protein (g/d) | 56.4±14.2 | 74.9±9.2 | 18.5±15.6 | 73.6±27.1 | 68.9±8.3 | -4.69±29.1 | 0.015 | 0.087 | <.001** | 0.001** |
| Cholesterol (mg/d) | 225.2±124.3 | 253.5±62,1 | 28.33±148.5 | 380.7±199.1 | 237.7±112.5 | -143±186.1 | 0.001** | 0.066 | 0.227 | <.001** |
| Fiber (g/d) | 21.5±6.3 | 28.6±4.8 | 7.1±7.3 | 15.3±4.8 | 25.9±4.1 | 10.56±7.1 | 0.135 | 0.980 | <.001** | 0.135 |
| Sodium (mg/d) | 3029±1101 | 2017.7±533.1 | -1011±1188 | 3212±1457 | 1891±363.4 | -1321±1465 | 0.434 | 0.609 | <.001** | 0.434 |
| Vitamin A (ug RE/d) | 945.5±583.7 | 1178.2±287.7 | 232.6±540.3 | 750.4±359.4 | 1069±143.8 | 318.3±374 | 0.595 | 0.318 | 0.003** | 0.595 |
| Thiamin (mg/d) | 1.1±0.3 | 1.2±0.2 | 0.13±0.3 | 1.2±0.4 | 1.1±0.2 | -0.03±0.5 | 0.167 | 0.250 | 0.017* | 0.167 |
| Riboflavin (mg/d) | 1.0±0.3 | 1.2±0.2 | 0.29±0.4 | 1.1±0.4 | 1.3±0.2 | 0.13±0.5 | 0.186 | 0.882 | <.001** | 0.186 |
| Vitamin C (mg/d) | 115.1±81.4 | 169.8±48.0 | 54.64±90.7 | 86.8±80.6 | 171.3±32.1 | 84.48±90.7 | 0.299 | 0.772 | <.001** | 0.299 |
| Vitamin E (mg/d) | 13.2±4.0 | 14.4±2.6 | 1.17±4.5 | 17.0±13.2 | 13.5±1.7 | -3.46±13.1 | 0.234 | 0.036* | 0.262 | 0.04* |
| Niacin (mg/d) | 13.6±3.8 | 17.1±2.5 | 3.49±4.3 | 17.8±5.4 | 16±2.2 | -1.89±6.3 | 0.001** | 0.408 | <.001** | 0.001** |
| Calcium (mg/d) | 376,7±128.1 | 468.9±78.8 | 92.2±159.5 | 362.6±173.8 | 446.7±88.4 | 84.12±198 | 0.879 | 0.785 | 0.001** | 0.874 |
| Potassium (mg/d) | 2540±786.7 | 3741.3±604.8 | 1201±901.2 | 2392±570.7 | 3325±479.6 | 932.6±722.1 | 0.328 | 0.181 | <.001** | 0.328 |
| Phosphorus (mg/d) | 919.0±225.2 | 1210.4±163.6 | 291.3±262.3 | 941.3±336.0 | 1101±164.6 | 159.7±394.8 | 0.275 | 0.164 | <.001** | 0.161 |
| Iron (mg/d) | 13.0±3.9 | 16.4±2.3 | 3.43±4.3 | 15.5± | 15±2 | -0.45±8.2 | 0.121 | 0.431 | <.001** | 0.025* |

All values are presented as means ± SD. TK group = Traditional diet style based Korean group. WK group = Western diet style based Korean group.
P value[a], *t*-test between changes of TK group and WK group; P value[b-d], group, time and time* group interaction effect of linear mixed-effect models after adjusted for age, sex and body weight.
* indicates statistically significant values (* p<.05 , ** p <.01)

## 3. The K-Dash effect on metabolic syndrome

*Whole subject difference in metabolic syndrome indicators*

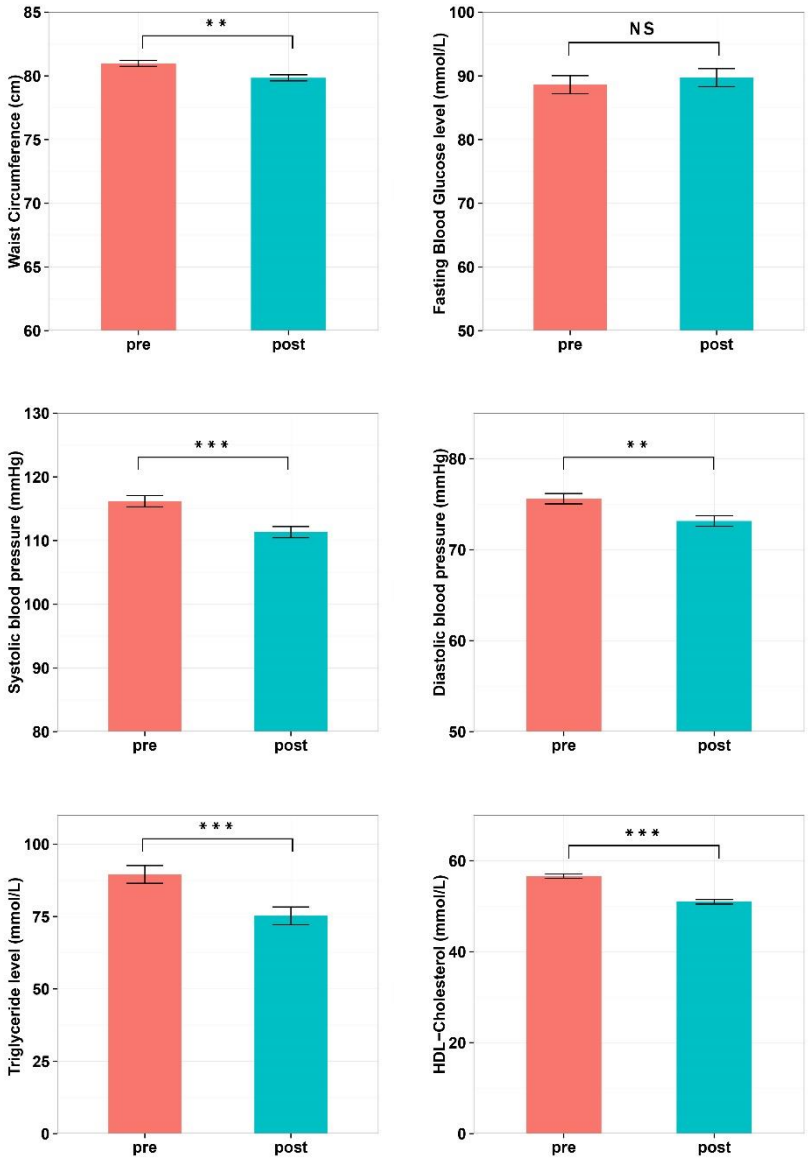
The effect of the 10-days K-DASH diet intervention on the anthropometric and metabolic variables in whole subjects is presented in **Table6.**

After 10-days intervention, whole subjects decreased in body weight and waist circumference (-0.90kg, -1.1cm respectively, $p<0.001$). Regarding to blood pressure, there were significant reduction in systolic blood pressure and diastolic blood pressure (-4.85mmHg, -2.45mmHg respectively, $p<0.01$). The level of triglyceride and HDL-cholesterol were significantly decreased (-14.45 mmol/L, -5.63mmol/L respectively, $p<0.001$); Fasting glucose level was slightly increased but it was not a significant change. (**Figure 1**)

*Whole subject difference in other biochemical variables.*

After 10-days intervention, whole subjects significantly decreased in total cholesterol level and LDL-cholesterol level (-14.45 mmol/L, -8.83 mmol/L, respectively, $p<0.001$). And the sodium concentration of Urine was significantly decreased. ($p<0.05$) No significant difference was observed in the level of hsCRP, HbA1C, Il-6, adiponectin. (**Table 6**)

**Figure 1.** The Change of metabolic syndrome indicators between pre-post K-DASH intervention




Metabolic syndrome indicators differences in whole subjects, at baseline and 10-days were analyzed using paired *t*-test.
* indicates statistically significant values (* p<.05 , ** p <.01, *** p<.001)

*Whole subject difference in insulin sensitivity and resistance*

After 10-days intervention, whole subjects significantly decreased in fasting insulin level and HOMA-IR insulin resistance index. (-2.30 pmol/L, -0.48 respectively, $p<0.01$) There was no significant difference in fasting glucose level. And insulin sensitivity index ISI, QUICKI both were increased after the intervention. (+0.30, +0.10 respectively, $p<0.001$) (**Figure 2**)

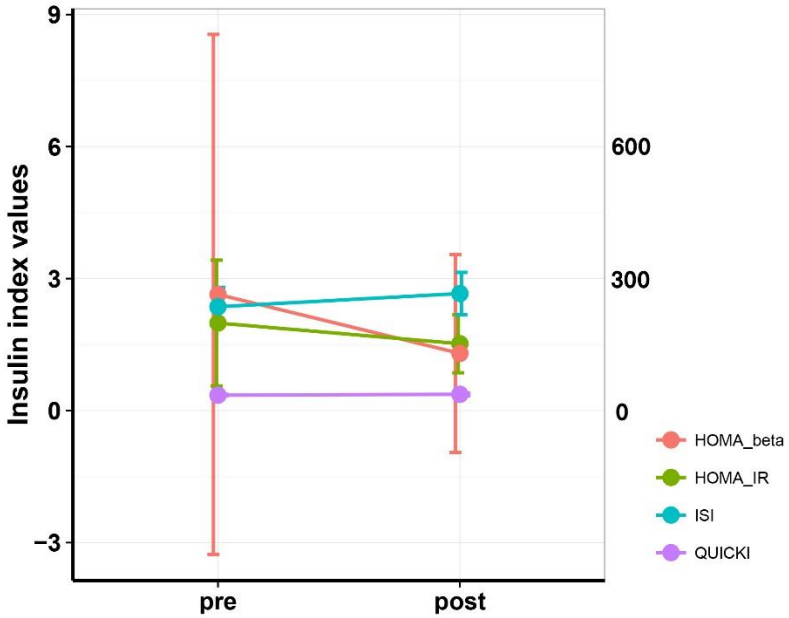



**Figure 2.** Changes in insulin sensitivity and resistance

Insulin sensitivity and resistance (HOMA_βcell, HOMA-IR, ISI ,QUICKI) differences in whole subjects, at baseline and 10-days were analyzed using paired *t*-test.

**Table 6.** Changes in anthropometric and metabolic variables in whole subjects

| | Whole subjects (n=65) | | | |
|---|---|---|---|---|
| | Baseline mean± SD | 10-days mean± SD | Mean difference | P-value[1] |
| Body weight (kg) | 59.7±12.2 | 58.8±12 | -0.9±0.8 | <.001** |
| BMI (kg/m$^2$) | 23.7±3.9 | 23.3±3.8 | -0.4±0.3 | <.001* |
| Waist (cm) | 81±9.2 | 79.8±9.1 | -1.1±2.7 | <.001** |
| Hip (cm) | 96.4±8 | 95.3±8 | -1±2.6 | 0.002** |
| Systolic BP (mmHg) | 116.2±14.8 | 111.3±13.9 | -4.8±10 | <.001* |
| Diastolic BP(mmHg) | 75.6±10.9 | 73.2±9.7 | -2.4±6.5 | 0.004** |
| Total cholesterol (mmol/L) | 181.9±31.4 | 167.4±26.7 | -14.4±18.4 | <.001 |
| LDL cholesterol (mmol/L) | 104.7±28.3 | 95.9±24.1 | -8.8±15.4 | <.001** |
| HDL- cholesterol (mmol/L) | 56.6±14.8 | 51±12.9 | -5.6±5.7 | <.001** |
| Triglyceride (mmol/L) | 89.5±47 | 75.3±48.6 | -14.3±34.9 | 0.002** |
| HbA1c | 5.6±0.7 | 5.6±0.7 | 0±0.2 | 0.564 |
| hsCRP | 1.4±2.7 | 1±2.3 | -0.4±2 | 0.109 |
| Adiponectin | 9860.4±6016.7 | 10059±9170.8 | 198.7±8274 | 0.847 |
| Interluekin-6 | 1.8±1.9 | 1.7±1.8 | -0.1±0.91 | 0.466 |
| Apolipoprotein A-I (g/L) | 157.9±26 | 142.7±23.8 | -15.2±13.4 | <.001** |
| Apolipoprotein B (g/L) | 91.5±23.1 | 85.4±19.8 | -6.1±14.3 | 0.001** |
| Fasting plasma glucose (mmol/L) | 88.6±28.1 | 89.7±17.2 | 1.1±16.2 | 0.582 |
| Fasting serum insulin (pmol/L) | 9±5.5 | 6.7±2.4 | -2.3±4.6 | <.001** |
| HOMA_IR | 1.99±1.43 | 1.52±0.66 | -0.48±1.20 | 0.002** |
| HOMA_β-cell | 264.2±591.4 | 129.8±225.5 | -134.4±546.1 | 0.06 |
| ISI | 2.36±0.44 | 2.66±0.48 | 0.30±0.39 | <.001** |
| QUICKI | 0.35±0.02 | 0.37±0.03 | 0.01±0.03 | <.001** |

All values are presented as means ± SD. * indicates statistically significant values (* p<.05 , ** p <.01)
[1]Paired Student's *t*-test was used for comparison baseline versus 10-days in dietary intakes.

*Between group difference in metabolic syndrome indicators*

**Table7** shows the effect of the 10-days K-DASH diet intervention on the anthropometric and metabolic variables by group.

In both intervention group, Body weight, BMI and waist circumference were significantly decreased ($p<0.05$); The changes of Body weight and BMI were much higher in WK group than TK group. (-1.29kg, -2.27cm respectively, $p<0.05$) Regarding to blood pressure, systolic blood pressure and diastolic blood pressure both were significantly decreased in TK group($p<0.01$); In WK group there was significant reduction in systolic blood pressure only ($p<0.05$). In both group, the level of HDL-cholesterol was significantly decreased (-4.17 mmol/L, -9.16 mmol/L respectively, $p<0.01$); TG level was significantly decreased only TK group ($p<0.01$). No significant difference was observed in fasting glucose level in both intervention group. A significant group difference in changes were observed in body weight, BMI, HDL-cholesterol; Interaction between time and group was observed in body weight, waist circumference and HDL-cholesterol ($p<0.05$).

*Between group difference in other biochemical variables.*

Compared to baseline, the levels of total cholesterol and LDL-cholesterol were significantly decreased in both group ($p<0.01$). And the level of Apolipoprotein A-1,B were significantly decreased in both group ($p<0.05$); Adiponectin level was increased in TK group and decreased in WK group. No significant difference was observed in the level of hsCRP, HbA1C, Il-6, adiponectin. A

significant group difference in changes were observed in adiponectin and Apolipoprotein A-I. Interaction between time and group was observed in Apolipoprotein A-I ($p<0.05$). (**Table 7**)

*Between group difference in insulin sensitivity and resistance*

**Table7** Shows insulin sensitivity and resistance changes by group. After 10-days intervention, there were significantly reduction in fasting insulin level and HOMA-IR insulin resistance index in both group. ($p<0.05$) HOMA-β cell index was significantly decreased in TK group (-50.8, $p<0.01$). And insulin sensitivity index ISI, QUICKI were significantly increased in both group after the intervention. ($p<0.05$) A significant pre-post effect was observed in HOMA-IR, HOMA- β cell and QUICKI; Interaction between time and group was observed in QUICKI index ($p<0.05$).

**Table 7.** Changes in anthropometric and metabolic variables between baseline and 10-days intervention by group

| | TK group (n=46) | | | WK group (n=19) | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Baseline mean± SD | 10-days mean± SD | Mean difference | Baseline mean± SD | 10-days mean± SD | Mean difference | P value[a] | P value[b] | P value[c] | P value[d] |
| Sex (female, n %) | 46 (100) | | | 8(42.1) | | | | | | |
| Age (y) | 50.1±6.67 | | | 29.4±5.02 | | | | | | |
| Body weight (kg) | 56.8±7.81 | 56.04±7.7 | -0.73(0.77)** | 66.65±17.5 | 65.36±17.1 | -1.29(0.87)** | 0.014* | 0.017* | <.001** | 0.014* |
| BMI(kg/m$^2$) | 23.16±2.69 | 22.79±2.68 | -0.36±0.30** | 25.15±5.66 | 24.6±5.53 | -0.55±035** | 0.032* | 0.011* | <.001** | 0.032* |
| Waist (cm) | 80.8±7.13 | 80.17±7.3 | -0.67±1.97* | 81.31±13.2 | 79.04±12.7 | -2.27±3.84* | 0.099 | 0.083 | 0.089 | 0.03* |
| Hip (cm) | 94.6±4.71 | 93.6±4.61 | -1.01±1.88** | 100.5±11.9 | 99.4±12.2 | -1.13±3.83 | 0.899 | 0.017* | 0.01** | 0.868 |
| Systolic BP (mmHg) | 115.8±15.4 | 110.8±14.3 | -4.95±11.1** | 117.1±13.5 | 112.5±13.2 | -4.58±7.06* | 0.870 | 0.710 | 0.001** | 0.891 |
| Diastolic BP(mmHg) | 74.6±9.19 | 71.6±8.09 | -2.98±6.46** | 77.9±14.1 | 76.8±12.4 | -1.16±6.76 | 0.312 | 0.815 | 0.003** | 0.312 |
| TC (mmol/L) | 179.6±33.8 | 167.3±27.7 | -12.3±18.9** | 187.3±24.8 | 167.6±24.6 | -19.6±16.4** | 0.145 | 0.915 | <.001** | 0.145 |
| LDL-C (mmol/L) | 101.6±30.5 | 93.7±24.2 | -7.91±15.6** | 112.2±20.9 | 101.1±23.5 | -11.0±14.9** | 0.459 | 0.616 | 0.001** | 0.459 |
| HDL-C (mmol/L) | 55,4±14.2 | 51.2±12.4 | -4.17±4.63** | 59.4±16.2 | 50.3±14.4 | -9.16±6.62** | 0.001** | 0.874 | <.001** | 0.001** |
| TG (mmol/L) | 93.7±51.4 | 78.7±53.5 | -14.9±37.4** | 79.5±33.4 | 66.8±33.7 | -12.6±29.0 | 0.809 | 0.635 | 0.005** | 0.809 |
| HbA1c | 5.65±0.78 | 5.71±0.75 | 0.06±0.16* | 5.34±0.35 | 5.25±0.29 | -0.08±0.34 | 0.092 | 0.744 | 0.086 | 0.025* |
| hsCRP | 1.17±2.15 | 0.64±0.64 | -0.53±2.18 | 1.99±3.65 | 1.89±4.12 | -0.10±1.52 | 0.439 | 0.194 | 0.079 | 0.439 |
| Adiponectin | 9891±6335 | 11197±10421 | 1306±9568 | 9785±5326 | 7303±4022 | -2482±1947** | 0.013* | 0.620 | 0.281 | 0.093 |
| Interluekin-6 | 1.64±1.76 | 1.58±1.70 | -0.05±0.72 | 2.11±2.15 | 1.96±2.09 | -0.15±1.27 | 0.759 | 0.096 | 0.687 | 0.701 |
| ApoA-I (g/L) | 156.7±24.9 | 144.8±22.8 | -11.9±11.4** | 160.8±28.9 | 137.6±26.1 | -23.1±14.7** | 0.002** | 0.985 | <.001** | 0.002* |
| Apo B (g/L) | 90.6±25.5 | 85.3±20.7 | -5.30±14.6* | 93.6±16.4 | 85.6±17.8 | -8.05±13.6* | 0.485 | 0.792 | 0.015* | 0.485 |
| FBG (mmol/L) | 92.9±31.3 | 93.7±17.6 | 0.74±16.4 | 78.1±13.5 | 80.0±11.4 | 2.00±15.8 | 0.777 | 0.917 | 0.759 | 0.777 |
| FSI (pmol/L) | 8.63±5.63 | 6.78±2.22 | -1.85±4.84* | 10.0±5.32 | 6.64±2.9 | -3.40±3.87** | 0.218 | 0.726 | 0.008** | 0.218 |

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| HOMA_IR | 1.99±1.46 | 1.58±0.62 | -0.41±1.25* | 2.00±1.37 | 1.36±0.73 | -0.64±1.01* | 0.491 | 0.851 | 0.024* | 0.494 |
| HOMA_β-cell | 143.6±85.6 | 92.8±41.1 | -50.8±69.1** | 556.3±1048 | 219.5±405.7 | -336.7±994 | 0.226 | 0.113 | 0.521 | 0.054 |
| ISI | 2.38±0.45 | 2.63±0.45 | 0.25±0.39** | 2.33±0.44 | 2.74±0.55 | 0.41±0.39** | 0.150 | 0723 | <.001** | 0.151 |
| QUICKI | 0.35±0.02 | 0.36±0.02 | 0.01±0.02* | 0.35±0.02 | 0.38±0.04 | 0.02±0.03** | 0.063 | 0.478 | 0.044* | 0.022* |

All values are presented as means ± SD. TK group = Traditional diet style based Korean group. WK group = Western diet style based Korean group.
P value[a], *t*-test between changes of TK group and WK group; P value[b-d], group, time and time* group interaction effect of linear mixed-effect models after adjusted for age sex.
* indicates statistically significant values (* p<.05 , ** p <.01)

# 4. The K-DASH effect on the intestinal microbiota

*Characterization of intestinal microbiota*

A total of 2,678,160 sequences were generated from 65 subjects stool sample. After quality filtering 2,573,832 sequences were obtained and the averages of 19,798 ± 7,100 sequences were detected per sample. **Figure3** shows differences in intestinal microbiota diversity between TK group and WK group at baseline.

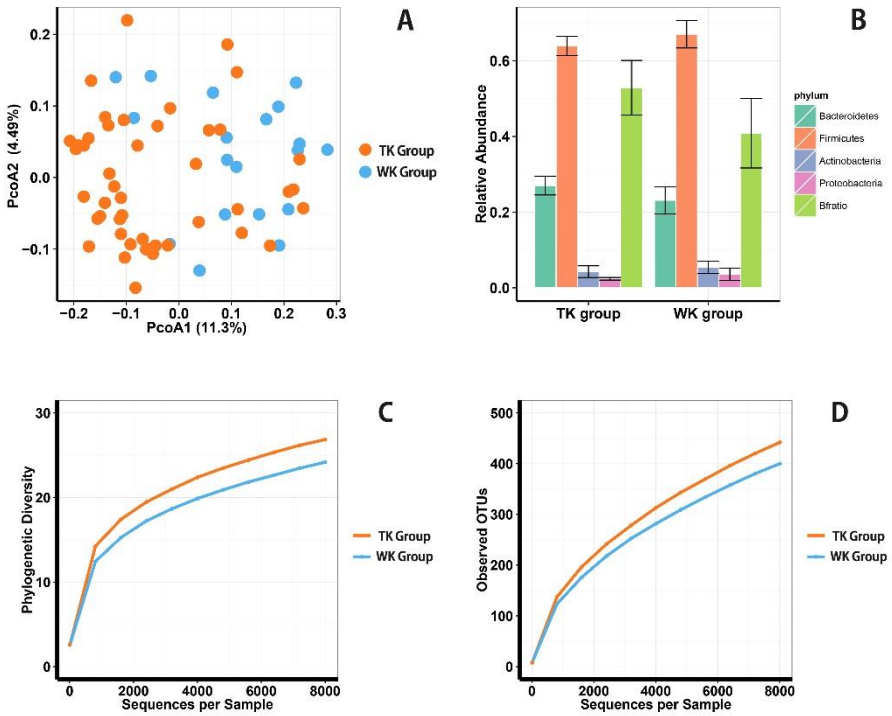



**Figure 3.** Characterization of gut microbiota communities by group

(A) Principal coordinate analysis (PCoA) of the intestinal microbiota communities in the TK group (orange spots) and WK group (blue spots) individuals at baseline. PcoA1 and PcoA2 values were estimated 11.3% and 4.49% respectively.

(B) Intestinal microbiota composition at phylum level box plot, in the TK, WK group.

(C) Rarefaction curves calculated for phylogenetic diversity (D) observed OTU units.

The alpha diversity of TK group intestinal microbiota was higher than WK group.(**Fig.3C,D**) In PcoA of both unweighted and weighted, based on phylogenetic analysis, indicated no clear separation between two group.(**Fig. 3A**) The composition of the phylum *Bacteroidetes* in TK group (27.0% ± 16.9%) was higher than that of WK group (22.1% ± 15.4%). In contrast, the composition of the phylum *Firmicutes* in WK group (68.2% ± 15.8%) was higher than that of TK group (63.9% ± 17.3%). The overall ratio of *Firmicutes* to *Bacteroidetes* (F/B ratio) was higher in WK group than TK group (2.37, 3.08 respectively), but it was not a significant differences. At genus level, *Faecalibacterium*, *Bacteroides* and *Prevotella* were predominated in the both group.

**Table 8.** Characterization of baseline gut microbiota communities by group

| | Relative contribution[1] (%) | | Group difference | |
|---|---|---|---|---|
| | TK group | WK group | Difference | P-value[2] |
| **Phylum level** | | | | |
| Bacteroidetes | 27.1±16.9 | 22.1±15.5 | 4.93±16.49 | 0.289 |
| Firmicutes | 64±17.4 | 68.2±15.9 | -4.24±16.92 | 0.352 |
| Actinobacteria | 4.3±10.6 | 5.5±6.8 | -1.21±9.6 | 0.064 |
| Proteobacteria | 2.4±2.4 | 3.5±6.8 | -1.06±4.15 | 0.861 |
| F/B ratio | 2.37±1.03 | 3.08±1.03 | -0.86±1.03 | 0.2398 |
| **Genus level** | | | | |
| Bacteroides | 10.4±10.59 | 15.24±10.82 | -4.84±10.66 | 0.039* |
| Prevotella | 11.18±15.17 | 3.37±9.71 | 7.81±13.84 | 0.003** |
| Enterococcus | 0.02±0.08 | 0.01±0.02 | 0.02±0.07 | 0.029* |
| Lactobacillus | 0.91±3.76 | 0.12±0.44 | 0.78±3.19 | 0.169 |
| Clostridium | 2.72±6.16 | 0.34±0.94 | 2.37±5.23 | 0.003** |
| Dorea | 1.1±0.94 | 1.75±1.5 | -0.65±1.13 | 0.177 |
| Bifidobacterium | 3.75±10.01 | 5.03±6.73 | -1.28±9.19 | 0.094 |
| Ruminococcus | 2.93±3.18 | 5.42±5.12 | -2.49±3.83 | 0.036* |
| Faecalibacterium | 9.19±7.31 | 17.62±8.49 | -8.43±7.66 | 0.001** |
| Butyricimonas | 0.11±0.15 | 0.05±0.09 | 0.06±0.14 | 0.049* |
| Roseburia | 0.22±0.43 | 0.35±0.43 | -0.14±0.43 | 0.177 |
| Streptococcus | 0.47±0.72 | 1.26±1.64 | -0.79±1.07 | 0.068 |
| Eubacterium | 2.91±5.44 | 0.93±1.38 | 1.97±4.66 | 0.239 |

[1] All values are presented as the average relative abundance(%) and SD. TK group = Traditional diet style based Korean group. WK group = Western diet style based Korean group.

[2] p-value were determined two-sided Mann-Whitney U test between baseline microbiota communities of two intervention group.

* indicates statistically significant values (* $p<.05$ , ** $p<.01$)

*The changes of relative abundance and diversity in the intestinal microbiota after 10-days K-DASH intervention.*

After 10-days K-DASH intervention, the microbiota diversity and composition of whole subjects were altered as presented in **Figure4**. The alpha diversity of whole subjects was increased after the intervention. (**Fig.4C,D**) The levels of *Bacteroidetes* was increased and *Firmicutes* was decreased. (+0.98%, -0.7% respectively, p=NS). The F/B ratio was decreased from 2.55 to 2.43, but it was not a significant differences.(**Table 9**)

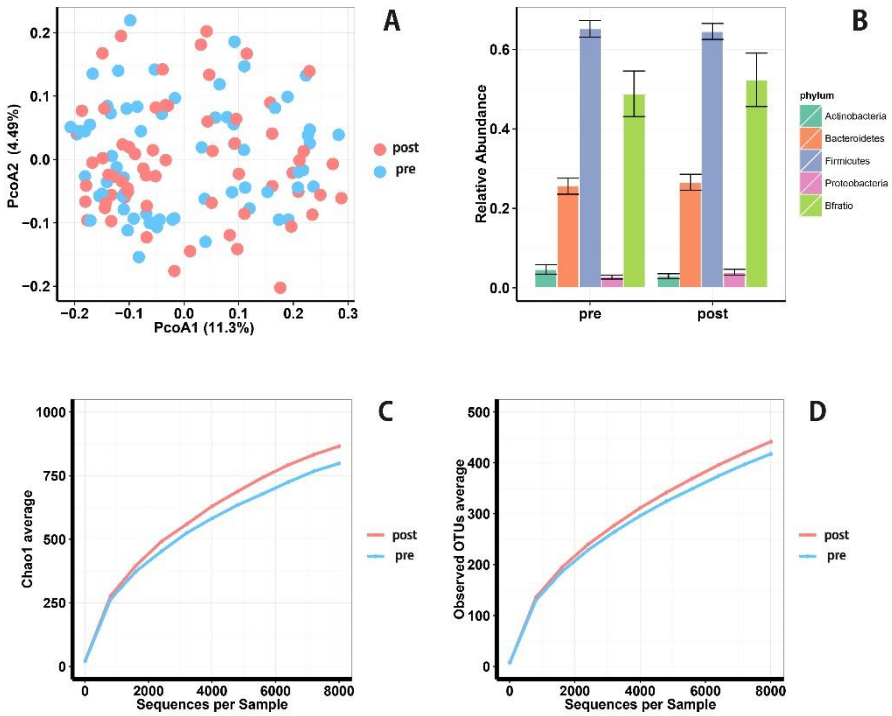



**Figure 4.** (A) Principal coordinate analysis (PCoA) of the intestinal microbiota communities in pre-intervention (blue spots) and post-intervention (red spots) individuals. PcoA1 and PcoA2 values were estimated 11.3% and 4.49% respectively.
(B) Intestinal microbiota composition at phylum level box plot, at pre-post intervention.
(C) Rarefaction curves calculated using chao1 (D) observed OTUs.

At genus level, the abundance of *Enterococcus, Lactococcus, Coprococcus, Lachnospria and Citrobacter* was significantly increased ($p<0.05$) and *Provotella* was increased but it was not a significant differences. In addition, there were significant reduction in *Dorea, Ruminococcus, Dialister and Parabacteroides* ($p<0.05$); *Bifidobacterium, Roseburia, Faecalibacterium* were slightly reduced but it was not a significant differences. (**Table 9**) At species level, the relative abundance of *B. plebeius, R. gnavus* and *E. biforme* was decreased and *Anaerostipes spp, Enterococcus spp, C. eutactus, P. nanceiensis* was increased ($p<0.05$)

**Table 9.** The changes in gut microbiota communities after 10-days K-DASH intervention – phylum, genus level

| | Relative contribution[1] (%) | | | |
|---|---|---|---|---|
| | Pre | Post | Difference | P-value[2] |
| **Phylum level** | | | | |
| Bacteroidetes | 25.58±16.52 | 26.56±16.27 | 0.98±17.25 | 0.658 |
| Firmicutes | 65.18±16.91 | 64.49±16.08 | -0.7±17.31 | 0.854 |
| Actinobacteria | 4.62±9.54 | 2.95±4.74 | -1.68±9.95 | 0.101 |
| Proteobacteria | 2.7±4.15 | 3.93±5.76 | 1.24±6.4 | 0.326 |
| F/B ratio | 2.55±1.02 | 2.43±0.99 | -0.71±1.00 | 0.814 |
| **Genus level** | | | | |
| Bacteroides | 11.82±10.81 | 11.63±10.42 | -0.2±10.31 | 0.859 |
| Prevotella | 8.91±14.19 | 10.23±15.99 | 1.33±10.98 | 0.988 |
| Enterococcus | 0.02±0.08 | 0.11±0.44 | 0.09±0.38 | 0.011 |
| Lactobacillus | 0.68±3.19 | 0.51±2.06 | -0.18±1.4 | 0.121 |
| Clostridium | 2.03±5.3 | 1.75±4.71 | -0.28±2.31 | 0.715 |
| Dorea | 1.29±1.17 | 0.98±0.85 | -0.32±0.9 | 0.012 |
| Bifidobacterium | 4.13±9.14 | 2.36±3.91 | -1.78±9.21 | 0.551 |
| Ruminococcus | 3.66±3.98 | 2.31±2.25 | -1.36±3.39 | 0.004 |
| Faecalibacterium | 11.66±8.53 | 11.58±8.6 | -0.09±9.39 | 0.546 |
| Butyricimonas | 0.1±0.14 | 0.13±0.25 | 0.03±0.24 | 0.859 |
| Roseburia | 0.26±0.44 | 0.24±0.34 | -0.03±0.54 | 0.870 |
| Streptococcus | 0.71±1.12 | 0.88±1.81 | 0.17±1.77 | 0.483 |
| Eubacterium | 2.34±4.71 | 1.83±3.96 | -0.51±3.18 | 0.066 |

[1] All values are presented as the average relative abundance(%) and SD. TK group = Traditional diet style based Korean group. WK group = Western diet style based Korean group.
[2] p-value were determined two-sided Mann-Whitney U test between baseline microbiota communities of two intervention group.
* indicates statistically significant values (* $p<.05$ , ** $p<.01$)

*Between group difference in microbiota communities at pre-post intervention*

The overall alpha diversity of TK group intestinal microbiota was higher than WK group at pre-post intervention. (**Figure 4**) And both group has creased in phylogenetic diversity after 10-days K-DASH diet intervention.

In phylum level, the abundance of *Bacteroidetes* was increased and the abundance of *Firmicutes* and F/B ratio was decreased in both group. The abundance of *Actinobacteria* was significantly decrease in WK group (-3.5%, $p<0.001$) In genus level, the relative abundance of *Bifidobacterium*, *Lactobacillus*, *Dialister* and *Dorea* was decreased only in WK group ($p<0.05$); the abundance of *Coprococcus*, *Anaerostipes*, *Sutterella* and *Desulfovibrio* was increased only in WK group ($p<0.05$). In TK group, *Parabacteroide* and *Ruminococcus* was decreased and *Enterococcus*, *Lactococcus*, *Streptococcus* and *Citrobacter* was significantly increased ($p<0.05$). In genus level, the abundance of *Bifidobacterium* spp, *Lactobacillus* spp, B.*adolescentis* L. *ruminis* and R.*gnavus* was significantly decreased only in WK group ($p<0.05$); the abundance of *Anaerostipes* spp, *Coprococcus* sp and E.*biforme* was increased in WK group ($p<0.05$). In TK group, the abundance of Enterococcus spp, R.*faecis*, C.*eutactus* and P.*nanceiensis* were increased and B.*plebeius*, R. *gnavus* was decreased ($p<0.05$) (**Table 10**)

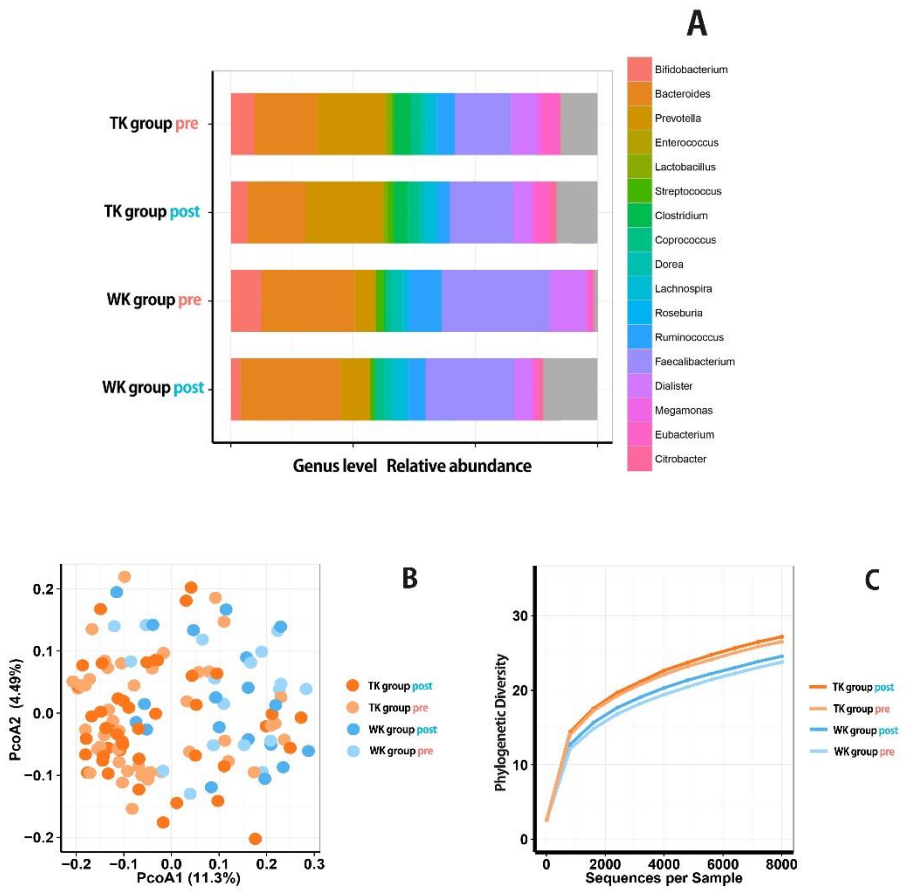



**Figure 5.** Comparison of microbiota communities changes by group

(A) Principal coordinate analysis (PCoA) of the intestinal microbiota communities in the TK group (orange spots) and WK group (blue spots) individuals at baseline. PcoA1 and PcoA2 values were estimated 11.3% and 4.49% respectively.

(B) Intestinal microbiota composition at phylum level box plot, in the TK, WK group.

(C) Rarefaction curves calculated for phylogenetic diversity (D) observed

**Table 10.** Comparison of gut microbiota communities changes by group - Phylum, Genus, Species level

| | TK group (n=46) | | | | WK group (n=19) | | | |
|---|---|---|---|---|---|---|---|---|
| | Baseline mean± SD | 10-days mean± SD | Mean difference | p-value[1] | Baseline mean± SD | 10-days mean± SD | Mean difference | p-value[2] |
| **Phylum level** | | | | | | | | |
| Bacteroidetes | 27.03±16.9 | 27.37±16.17 | 0.35±16.61 | 0.809 | 22.1±15.44 | 25.85±16.93 | 3.76±18.88 | 0.768 |
| Firmicutes | 63.94±17.34 | 62.91±15.21 | -1.03±15.89 | 0.743 | 68.18±15.84 | 67.23±18.17 | -0.95±20.84 | 0.922 |
| Actinobacteria | 4.27±10.54 | 3.37±4.97 | -0.9±11.31 | 0.58 | 5.47±6.73 | 1.92±4.05 | -3.56±5.19 | 0.001 |
| Proteobacteria | 2.39±2.38 | 4.02±6.49 | 1.64±6.79 | 0.851 | 3.45±6.79 | 3.71±3.58 | 0.26±5.39 | 0.157 |
| F/B ratio | 2.37±1.03 | 2.3±0.95 | -0.07±0.96 | 0.877 | 3.09±1.03 | 2.61±1.08 | -0.49±1.11 | 0.953 |
| **Genus level** | | | | | | | | |
| Methanobrevibacter | 0.73±3.98 | 0.49±3.22 | -0.25±0.98 | 0.389 | 0.02±0.06 | 0.02±0.04 | -0.01±0.03 | 0.641 |
| Bifidobacterium | 3.76±10.01 | 2.68±3.97 | -1.09±10.39 | 0.508 | 5.04±6.74 | 1.58±3.73 | -3.47±5.28 | 0.001 |
| Collinsella | 0.15±0.67 | 0.39±1.88 | 0.25±1.99 | 0.623 | 0.06±0.16 | 0.01±0.01 | -0.06±0.16 | 0.125 |
| Bacteroides | 10.41±10.6 | 9.65±9.57 | -0.76±10.47 | 0.818 | 15.25±10.83 | 16.4±11.08 | 1.16±10.04 | 0.542 |
| Parabacteroides | 1.86±1.52 | 1.22±1.41 | -0.64±10.47 | 0.01 | 2.22±3.7 | 1.44±1.43 | -0.79±3.58 | 0.766 |
| Prevotella | 11.19±15.18 | 12.47±17.11 | 1.29±10.47 | 0.936 | 3.38±9.72 | 4.79±11.56 | 1.41±11.48 | 0.891 |
| Butyricimonas | 0.12±0.15 | 0.16±0.28 | 0.04±10.47 | 0.98 | 0.06±0.1 | 0.06±0.12 | 0.01±0.06 | 0.846 |
| Odoribacter | 0.14±0.17 | 0.17±0.22 | 0.03±10.47 | 0.607 | 0.15±0.19 | 0.21±0.37 | 0.07±0.38 | 0.669 |
| Paraprevotella | 0.47±0.75 | 0.32±0.43 | -0.15±10.47 | 0.238 | 0.07±0.21 | 0.1±0.3 | 0.03±0.16 | 0.813 |
| Enterococcus | 0.03±0.09 | 0.15±0.52 | 0.13±10.47 | 0.026 | 0.01±0.03 | 0.02±0.04 | 0.01±0.05 | 0.297 |
| Lactobacillus | 0.91±3.77 | 0.71±2.42 | -0.2±10.47 | 0.631 | 0.13±0.45 | 0.03±0.07 | -0.11±0.38 | 0.004 |
| Lactococcus | 0.01±0.01 | 0.15±0.81 | 0.15±10.47 | 0.006 | 0.02±0.03 | 0.12±0.18 | 0.1±0.18 | 0.007 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Streptococcus | 0.48±0.72 | 0.95±2.03 | 0.48±10.47 | 0.029 | 1.27±1.65 | 0.7±1.14 | -0.57±1.33 | 0.088 |
| Clostridium | 2.72±6.16 | 2.4±5.49 | -0.33±10.47 | 0.768 | 0.35±0.94 | 0.19±0.2 | -0.17±0.8 | 0.953 |
| Anaerostipes | 0.37±0.5 | 0.54±0.92 | 0.17±10.47 | 0.287 | 0.26±0.27 | 1.05±1.72 | 0.8±1.67 | 0.055 |
| Blautia | 1.35±0.98 | 1.45±0.99 | 0.11±10.47 | 0.752 | 2.12±2.34 | 2.22±1.58 | 0.1±2.44 | 0.490 |
| Coprococcus | 1.45±1.4 | 1.83±1.5 | 0.38±10.47 | 0.086 | 0.99±0.66 | 1.47±1.12 | 0.49±0.82 | 0.019 |
| Dorea | 1.1±0.95 | 0.84±0.57 | -0.26±10.47 | 0.086 | 1.75±1.51 | 1.31±1.26 | -0.45±0.84 | 0.033 |
| Lachnospira | 1.49±1.98 | 1.87±1.89 | 0.38±10.47 | 0.65 | 0.65±1.43 | 2.35±3.91 | 1.71±4.19 | 0.006 |
| Roseburia | 0.22±0.44 | 0.26±0.38 | 0.04±10.47 | 0.391 | 0.36±0.44 | 0.2±0.22 | -0.16±0.47 | 0.396 |
| Ruminococcus | 2.93±3.18 | 2.07±2 | -0.86±10.47 | 0.026 | 5.42±5.12 | 2.88±2.73 | -2.55±5.1 | 0.067 |
| Faecalibacterium | 9.2±7.31 | 10.37±8.24 | 1.18±10.47 | 0.711 | 17.63±8.49 | 14.5±8.98 | -3.13±9.55 | 0.157 |
| Oscillospira | 0.59±0.58 | 0.64±0.74 | 0.05±10.47 | 0.826 | 0.4±0.39 | 0.62±0.81 | 0.22±0.63 | 0.211 |
| Acidaminococcus | 0.49±2.35 | 0.34±1.93 | -0.16±10.47 | 0.09 | 0.68±2.09 | 0.53±2.28 | -0.16±1.21 | 0.966 |
| Dialister | 4.27±6.5 | 3.05±4.19 | -1.22±10.47 | 0.219 | 6.05±9.46 | 2.94±4.24 | -3.11±6.61 | 0.037 |
| Megamonas | 0.84±3.96 | 0.48±1.76 | -0.37±10.47 | 0.134 | 0.18±0.51 | 0.36±1.33 | 0.19±1 | 0.427 |
| Megasphaera | 0.26±0.88 | 0.23±1.08 | -0.03±10.47 | 0.25 | 2.5±9.21 | 0.61±2.29 | -1.89±6.95 | 0.415 |
| Phascolarctobacterium | 2.48±3.87 | 1.4±2.3 | -1.08±10.47 | 0.173 | 0.85±1.36 | 2.05±2.74 | 1.21±1.77 | 0.014 |
| Catenibacterium | 1.82±3.82 | 1.28±2.87 | -0.55±10.47 | 0.254 | 0.02±0.03 | 0.01±0.02 | -0.01±0.02 | 0.147 |
| Eubacterium | 2.91±5.45 | 2.31±4.54 | -0.6±10.47 | 0.211 | 0.94±1.39 | 0.66±1.48 | -0.29±1.76 | 0.134 |
| Sutterella | 0.47±0.71 | 0.34±0.45 | -0.13±10.47 | 0.181 | 0.26±0.38 | 0.47±0.46 | 0.22±0.51 | 0.031 |
| Bilophila | 0.22±0.34 | 0.13±0.16 | -0.1±10.47 | 0.094 | 0.19±0.25 | 0.19±0.25 | 0.01±0.2 | 0.933 |
| Desulfovibrio | 0.26±0.72 | 0.14±0.32 | -0.13±10.47 | 0.521 | 0.24±0.69 | 0.13±0.36 | -0.12±0.39 | 0.027 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Campylobacter | 0.01±0.01 | 0.01±0.01 | 0.01±10.47 | 0.653 | 0.01±0.01 | 0.01±0.03 | 0.01±0.03 | 0.688 |
| Citrobacter | 0.16±0.34 | 1.3±3.71 | 1.14±10.47 | 0.001 | 0.16±0.42 | 0.86±2.03 | 0.7±2.09 | 0.067 |
| Haemophilus | 0.25±0.57 | 0.27±0.83 | 0.03±10.47 | 0.313 | 0.11±0.16 | 0.1±0.26 | -0.01±0.24 | 0.922 |
| **Species lvel** | | | | | | | | |
| Bifidobacterium sp | 0.15±0.26 | 0.18±0.39 | 0.04±0.43 | 0.881 | 0.48±0.8 | 0.07±0.12 | -0.41±0.8 | 0.003 |
| Bifidobacterium *adolescentis* | 3.61±9.82 | 2.49±3.63 | -1.12±10.1 | 0.543 | 4.54±6.64 | 1.5±3.67 | -3.04±5.08 | 0.001 |
| Collinsella *aerofaciens* | 0.15±0.67 | 0.39±1.88 | 0.25±1.99 | 0.623 | 0.06±0.16 | 0.01±0.01 | -0.06±0.16 | 0.125 |
| Bacteroides sp | 7.73±8.63 | 6.97±8.01 | -0.76±8.46 | 0.522 | 9.7±7.81 | 10.84±9.8 | 1.14±7.81 | 0.768 |
| Bacteroides *coprophilus* | 0.7±1.72 | 0.77±1.94 | 0.08±0.74 | 0.619 | 0.18±0.77 | 0.63±2.71 | 0.45±1.95 | 0.653 |
| Bacteroides *eggerthii* | 0.05±0.15 | 0.06±0.17 | 0.02±0.16 | 0.496 | 0.56±2.25 | 0.01±0.03 | -0.55±2.25 | 0.563 |
| Bacteroides *fragilis* | 0.36±0.67 | 0.38±0.66 | 0.02±0.86 | 0.368 | 1.04±1.29 | 0.96±1.43 | -0.09±1.49 | 0.922 |
| Bacteroides *ovatus* | 0.11±0.27 | 0.13±0.42 | 0.02±0.2 | 0.945 | 0.71±1.17 | 0.52±0.89 | -0.2±0.74 | 0.088 |
| Bacteroides *plebeius* | 0.55±1.05 | 0.46±0.94 | -0.1±0.92 | 0.035 | 1.03±2.52 | 0.66±1.65 | -0.37±1.55 | 1 |
| Bacteroides *uniformis* | 0.88±1.36 | 0.87±1.19 | -0.02±1.64 | 0.885 | 1.91±2.55 | 2.69±3.24 | 0.78±3.02 | 0.374 |
| Prevotella sp | 0.84±3.92 | 0.45±1.68 | -0.39±2.41 | 0.756 | 0.2±0.67 | 0.56±1.64 | 0.36±1.19 | 0.422 |
| Prevotella *copri* | 10.11±14.5 | 11.84±16.97 | 1.74±10.39 | 0.877 | 2.92±9.78 | 4.19±11.67 | 1.27±11.39 | 0.739 |
| Prevotella *intermedia* | 0.01±0.01 | 0±0 | -0.01±0.01 | 1 | 0±0 | 0±0 | 0±0 | 0.36 |
| Prevotella *melaninogenica* | 0.01±0.01 | 0.01±0.01 | 0.01±0.01 | 0.36 | 0.01±0.02 | 0.01±0.01 | -0.01±0.02 | 0.952 |
| Prevotella *nanceiensis* | 0±0 | 0.01±0.01 | 0.01±0.01 | 0.004 | 0.01±0.01 | 0±0 | -0.01±0.01 | 0.5 |
| Prevotella *stercorea* | 0.25±0.59 | 0.19±0.4 | -0.07±0.47 | 0.216 | 0.26±1.12 | 0.05±0.19 | -0.22±0.94 | 0.375 |
| Enterococcus sp | 0.03±0.09 | 0.15±0.52 | 0.13±0.45 | 0.026 | 0.01±0.03 | 0.02±0.04 | 0.01±0.05 | 0.297 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Lactobacillus sp | 0.31±2.06 | 0.29±1.93 | -0.02±0.14 | 0.238 | 0.05±0.14 | 0.01±0.01 | -0.05±0.14 | 0.013 |
| Lactobacillus *brevis* | 0.01±0.01 | 0.01±0.01 | -0.01±0.01 | 0.25 | 0±0 | 0±0 | 0±0 | 0.25 |
| Lactobacillus *iners* | 0±0 | 0.01±0.01 | 0.01±0.01 | 0.25 | 0±0 | 0.01±0.01 | 0.01±0.01 | 1 |
| Lactobacillus *mucosae* | 0.1±0.53 | 0.06±0.41 | -0.04±0.16 | 0.098 | 0.01±0.01 | 0±0 | -0.01±0.01 | 1 |
| Lactobacillus *reuteri* | 0.14±0.95 | 0.01±0.01 | -0.14±0.95 | 0.547 | 0.01±0.02 | 0±0 | -0.01±0.02 | 0.25 |
| Lactobacillus *ruminis* | 0.33±0.66 | 0.36±0.79 | 0.03±0.87 | 0.913 | 0.06±0.23 | 0.02±0.07 | -0.04±0.17 | 0.032 |
| Lactobacillus *zeae* | 0.04±0.21 | 0.01±0.01 | -0.03±0.21 | 0.734 | 0.02±0.07 | 0±0 | -0.02±0.07 | 0.125 |
| Clostridium sp | 2.71±6.16 | 2.39±5.49 | -0.32±2.69 | 0.617 | 0.34±0.95 | 0.18±0.2 | -0.16±0.8 | 0.68 |
| Clostridium *butyricum* | 0.01±0.02 | 0.01±0.01 | -0.01±0.02 | 0.579 | 0.01±0.01 | 0.01±0.01 | -0.01±0.01 | 0.75 |
| Clostridium *perfringens* | 0.01±0.01 | 0.01±0.01 | 0.01±0.01 | 0.761 | 0.01±0.01 | 0.01±0.01 | -0.01±0.01 | 0.75 |
| Pseudoramibacter -Eubacterium sp | 0.01±0.02 | 0.01±0.01 | -0.01±0.02 | 0.562 | 0.01±0.03 | 0.01±0.01 | -0.01±0.03 | 0.625 |
| Anaerostipes sp | 0.37±0.5 | 0.54±0.92 | 0.17±0.87 | 0.287 | 0.26±0.27 | 1.05±1.72 | 0.8±1.67 | 0.055 |
| Coprococcus sp | 1.45±1.4 | 1.83±1.5 | 0.38±1.36 | 0.09 | 0.99±0.66 | 1.47±1.12 | 0.49±0.82 | 0.019 |
| Coprococcus *eutactus* | 0.001±0.001 | 0.002±0.001 | 0.001±0.001 | 0.0478 | 0.001±0.001 | 0.002±0.001 | 0.001±0.001 | 0.625 |
| Dorea *formicigenerans* | 0.49±0.49 | 0.42±0.32 | -0.08±0.44 | 0.407 | 0.55±1.04 | 0.47±0.97 | -0.08±0.36 | 0.396 |
| Roseburia *other* | 0.01±0.01 | 0.01±0.01 | 0.01±0.01 | 0.321 | 0.01±0.01 | 0.01±0.01 | -0.01±0.01 | 0.375 |
| Roseburia sp | 0.13±0.43 | 0.08±0.15 | -0.06±0.44 | 0.697 | 0.18±0.31 | 0.07±0.07 | -0.12±0.31 | 0.595 |
| Roseburia *faecis* | 0.09±0.14 | 0.18±0.29 | 0.09±0.3 | 0.06 | 0.18±0.25 | 0.14±0.21 | -0.04±0.27 | 0.378 |
| Ruminococcus *gnavus* | 2.63±2.98 | 1.84±1.88 | -0.79±2.09 | 0.022 | 4.84±4.96 | 2.32±2.25 | -2.52±4.76 | 0.026 |
| Faecalibacterium *prausnitzii* | 9.2±7.31 | 10.37±8.24 | 1.18±9.13 | 0.711 | 17.63±8.49 | 14.5±8.98 | -3.13±9.55 | 0.157 |
| Eubacterium sp | 0.01±0.01 | 0.01±0.01 | -0.01±0.01 | 0.375 | 0.01±0.01 | 0.01±0.01 | -0.01±0.01 | 0.25 |

| | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| Eubacterium *biforme* | 2.87±5.46 | 2.28±4.55 | -0.59±3.62 | 0.271 | 0.71±1.35 | 0.43±1.25 | -0.28±1.56 | 0.074 |
| Eubacterium *dolichum* | 0.05±0.11 | 0.04±0.1 | -0.01±0.12 | 0.101 | 0.23±0.4 | 0.23±0.46 | -0.01±0.27 | 0.747 |
| Fusobacterium sp | 0.01±0.08 | 0.00±0.00 | -0.01±0.08 | 0.303 | 0.11±0.42 | 0.02±0.04 | -0.09±0.38 | 0.588 |

[1] All values are presented as the average relative abundance(%) and SD. TK group = Traditional diet style based Korean group. WK group = Western diet style based Korean group.
[2] p-value[1] and p-value[2] were determined two-sided Wilcoxon signed rank test between pre-post intervention microbiota changes by group respectively.
* indicates statistically significant values (* p<.05, ** p <.01)

*Correlation between Metabolic variables and gut microbiota*

To assess the relationship between metabolic variables and gut microbiota changes, the Pearson correlation test were used. (**Figure 5**)

The changes of major phylum of gut microbiota *Bacteriodetes* and *Firmicutes* were negatively correlated across whole participants. ($r = -0.834$, $p<0.001$) Also, there was positive relationship between the changes of blood insulin level and the changes of F/B ratio, the abundances of *Lactobacillus*, *Lactobacillus reuteri*, *Lactobacillus zeae*, *Clostridium butyricum*, *Roseburia sp.* ($r=0.732$, $p<0.001$; $r=0.635$, $p<0.001$; $r=0.739$, $p<0.001$; $r=0.733$, $p<0.001$, $r=0.728$, $p<0.001$; $r=0.681$, $p<0.001$). Furthermore, regarding to insulin sensitivity and resistance, the positive relationship were observed between the changes of HOMA-IR index and the F/B ratio, the abundances of *Lactobacillus*, *Lactobacillus reuteri*, *Lactobacillus zeae*, *Clostridium butyricum*, *Roseburia sp.* ($r=0.699$, $p<0.001$; $r=0.522$, $p<0.001$; $r=0.739$, $p<0.001$; $r=0.695$, $p<0.001$, $r=0.697$, $p<0.001$; $r=0.651$, $p<0.001$). In contrast, the changes of Enterococcus abundance was negatively correlated with blood insulin level and HOMA-IR ($r = -0.668$, $p<0.001$; $r = -0.617$, $p<0.001$ respectively). The changes of Parabacteroides abundance was negatively correlated with HOMA-β cell function. ($r=-0.425$, $p<0.001$). And there was a positive relationship between the relative abundance of Clostridium sp and hsCRP changes. ($r=0.634$, $p<0.001$)

Regarding to MetS indicators, the changes of weight, BMI were correlated with the abundance of *Roseburia, Bifidobacterium sp, Collinsella aerofaciens, Prevotella melaninogenica, and Lactobacillus iners* but not strongly. ($|r| <0.5$, $p<0.05$) And the change of SBP was correlated with the abundance of

*Clostridium* and *Lactobacillus brevis*. (r=0.305, p=0.013; r= -0.286, p= 0.021) And there was positive relationship between the change of DBP and the abundance of *Clostridium*(r=0.311, p=0.012). Finally, the fasting blood glucose level was correlated with *Desulfovibrio* and *Bacteroides uniformis* abundance. (r=0.366, p=0.003; r= -0.321, p=0.009 respectively)

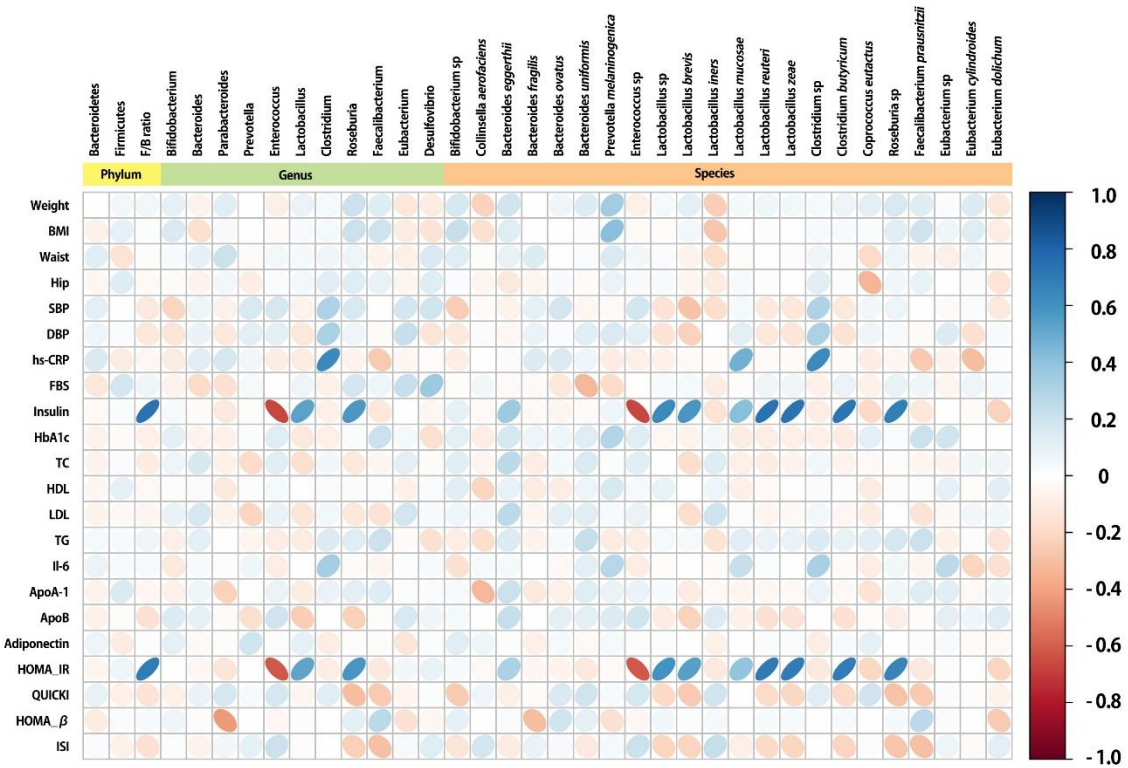



**Figure 6.** The changes of relative abundance of gut microbiota is correlated with metabolic syndrome indicators and insulin sensitivity and resistance indexes.

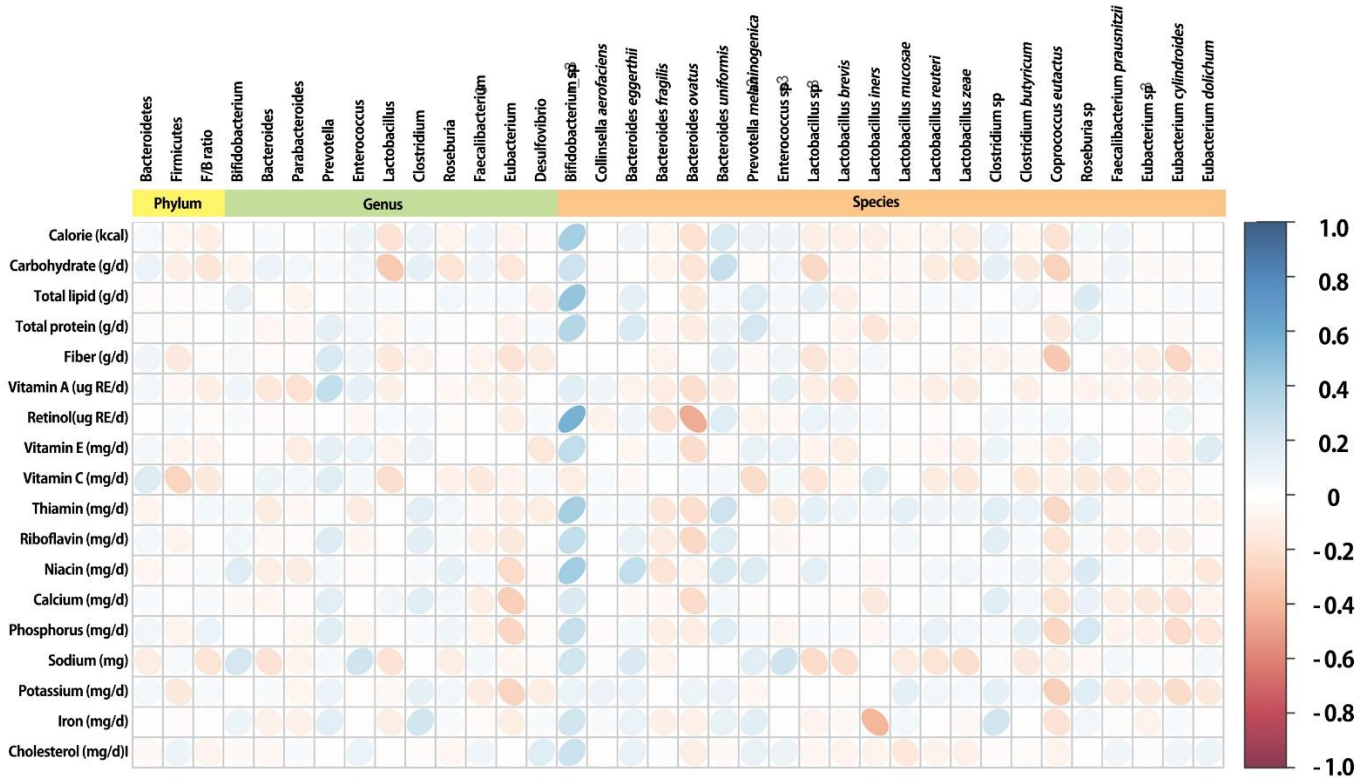



**Figure 7.** The overall correlation plot between the changes of nutrients intakes and gut microbiota abundance

# IV. DISCUSSION

This dietary intervention study conducted 10-days Korean DASH diet with traditional Korean diet based group and western diet based Korean and examined the effect on metabolic syndrome indicators and insulin sensitivity and resistance. The K-DASH diet that composed of rich in grains, vegetables, nuts and lower sodium content, improved risk factors for Mets including lowered waist circumference, systolic blood pressure and diastolic blood pressure and triglycerides level. These findings were similar to those studies reported that Korean traditional diets and DASH diet respectively improved the risk of metabolic syndrome.[6, 33, 34] However the beneficial effect of traditional Korean diet is controversial, because the traditional Korean diet includes high sodium and carbohydrates that could increase the risk of metabolic syndrome too.[35, 36] Thus, in this study the traditional diet was modified to low sodium contents and moderate carbohydrate composition based on DASH diet patterns. Therefore the effects of K-DASH diet were significant in both traditional diet style based Korean group (TK group) and western diet style based Korean group (WK group).

In this study with reduction of total cholesterol, the HDL-cholesterol level also was decreased. As is well known, high HDL-cholesterol level reduce the risk of cardiovascular heart disease.[37] Thus, the reduced HDL-cholesterol level could be a risk factor for metabolic syndrome. Similar findings were observed in other studies that restricted dietary fat intakes or weight loss reduction in HDL-cholesterol in the short time. [38, 39] However long term effect of weight loss on HDL-cholesterol was beneficial that those who reduced their weight

and maintained the body weight for a long time had shown higher HDL-cholesterol level.[40-42] Therefore, long term K-DASH diet is expected to recover or raise the HDL-cholesterol level and reduce the risk for metabolic syndrome and cardiovascular heart diseases.

K-DASH contained high fiber and protein, low fat and sodium that improves the gut microbiota composition and diversity resulting in lowering insulin resistance. A growing numbers[43, 44] studies have been reported the effects of DASH diet on HOMA-IR, insulin sensitivity and Hinderliter et al[45] reported that DASH eating plan improves insulin sensitivity and it is more effective when comprehensive lifestyle modification program implemented too. Previous studied reported that a high level of hs-CRP or IL-6 and lower level of adiponectin were associated with a greater risk of MetS. However in this study, there were no significant changes were observed in the inflammation markers such as hs-CRP, IL-6 and adiponectin.

The changes of dietary patterns affect the intestinal microbiota composition and the microbiota contributes to human health.[16] In this study, the relative abundance of *Bacteroidetes* was increased and *Firmicutes* was decreased after the intervention. Previous studies showed that the weight loss has been directly related to decreased *Firmicutes/Bacteroidetes* and in obese the proportion of *Firmicutes* was high and *Bacteroidetes* was low. [14] Likewise, this study showed the elevated *Bacteroidetes* and lowered *Firmicutes* abundance according to weight loss.

The dietary fiber is fermented into short-chain fatty acid (SCFA) by gut bacteria and the product, especially butyrate, have been studied for its role in modulating

inflammation and maintenance of gut barrier. [46] Contrary to expectation, the relative abundance of short-chain fatty acid producing bacteria (*Clostridium butyricum,, Faecalibacterium prausnitzii, Eubacterium sp ,Roseburia sp*) were reduced after the intervention, in spite of increased fiber intakes. Similar findings were observed in obese study that restricted their diet and another study that reduced carbohydrate intakes.[47, 48] It shows the SCFA producing *Eubacterium sp, Roseburia sp* are dependent on dietary carbohydrates to maintain their population in the intestine and the changes of dietary composition and relatively reduced whole carbohydrate ratio lead to reduction in short-chain fatty acid producing bacteria.

These results suggest that K-DASH diet could play an important role in the treatment of metabolic syndrome in Korean. However certain limitations exist. The sample size was small and a lack of a control group who intakes their usual diet during same intervention periods. In addition there was a limit to discover the precise effect mechanisms between diet, gut microbiota and human health. However, there was a difference in TK group and WK group regarding to body weight, BMI, HDL-cholesterol, adiponectin changes and interaction between time and group was observed in the changes of body weight, waist circumference and HDL-cholesterol. And the baseline intestinal microbiota composition and diversity was different. Thus, there is a possibility that previously formed gut microbiota can affect the efficacy of dietary intervention. Therefore prospective studies in large population are need to confirm whether the host's baseline microbiota composition predicts response and to establish the relationship between dietary modulation and gut microbiota and MetS improvement.

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## VI. Abstract in Korean (국문 초록)

# 한국식 DASH 식단이 대사증후군, 인슐린저항성 및 장내미생물 구성에 미치는 영향에 대한 연구

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대사증후군은 고혈압, 지질대사 및 내당능 장애 등의 위험요인이 복합적으로 나타나는 상태로, 한국인에게서 유병율이 증가하는 추세이다. 대사증후군은 생활습관과 밀접한 관련이 있으며, 특히 개인의 식습관과 관련이 깊다. 따라서 대사증후군의 예방 및 관리를 위해서는 한국인에게 맞는 건강한 식사패턴을 밝혀내는 것이 중요하다. 전통 한식은 다양한 곡류와 채소로 구성된 저지방 고식이섬유의 특성을 갖고 있어 대사증후군 등 만성질환에 긍정적인 효과를 낼 것으로 기대되나, 아직 그 근거가 부족한 상태이다. 이에 본 연구에서는 전통한식을 개량한 건강 한식 식단을 구성하고, 그 식단이 대사증후군 및 인슐린저항성 지표에 어떤 개선 효과를 보이는지 연구하였다.

본 연구를 위해 전통 한식을 섭취해 온 대상자 46 명과 해외에서 오래 거주하여 식사패턴이 전통한식과 다른 한국인 20 명을 모집하였다. 개량된 전통한식은 기존의 DASH 식단을 바탕으로 고식이섬유, 저 나트륨, 고단백질의 식품으로 구성하였고, 10 일동안 대상자들에게 제공되었다. 식단 섭취 전후에는 신체계측 및 소변, 혈액을 통한 생화학적 지표를 측정하였고, 수집된 자료를 이용하여 인슐린 저항성 지표를 산출하였다. 또한 대상자들의 장내미생물 변화에 대해 분석하기 위해 식단 섭취 전후에 각각 분변을 수집한 뒤 Illumina Miseq 을 이용하여 sequencing 데이터를 추출하였고, 장내미생물의 군집 및 다양성을 분석하였다.

참여 대상자들이 10 일동안 섭취한 한국식 DASH 식단은 기존에 대상자들이 섭취하던 식사 패턴과 차이가 있었다. 전반적인 단백질 및 식이섬유의 섭취량이 유의하게 증가했고, 나트륨 섭취량이 감소했다. 또한 비타민 A, B, C 군의 섭취량이 유의하게 증가하였다. 식사패턴의 변화로 인한 대사증후군 지표의 개선효과를 분석한 결과, 전체대상자에서 몸무게와 허리둘레가 유의하게 감소했으며 (-0.9kg, $p<0.001$; -1.1cm, $p<0.001$), 수축기 혈압과 이완기 혈압이 감소하였다. (-4.8mmHg, $p<0.001$; -2.4mmHg, $p<0.001$) 또한, 전반적인 콜레스테롤 수치가 감소하면서, HDL-콜레스테롤 수치와 중성지방(TG) 수치가 유의하게 감소하였다. (-5.6mmol/L, $p<0.001$; -14.3mmol/L, $p<0.05$) 공복혈당은 큰 변화가 없었으나, 인슐린 저항성 및 민감도 지표에서 개선되는 효과를 보였다. 인슐린 저항성 지표인 HOMA-IR 이 유의하게 감소하였고(-0.48, $p<0.002$) 인슐린 민감도 지표인 QUICKI 와 ISI 는 모두 유의하게 증가하였다. (+0.01, $p<0.001$; +0.3, $p<0.001$)

한국식 DASH 식단 섭취로 인한 장내미생물의 구성과 다양성을 분석한 결과, 개체 내 다양성 지표인 alpha diversity 가 증가하였다. 또한 문 단계에서 *Bacteroidetes* 의 상대적인 존재비는 증가하였고 *Firmicutes* 의 존재비는 감소하여 Firmicutes/*Bacteroidetes*

ratio 가 감소하였다. 속 단계에서는 *Enterococcus, Lactococcus, Coprococcus, Lachnospria* 그리고 *Citrobacter* 가 유의하게 증가했으며, *Dorea, Ruminococcus, Dialister* 그리고 *Parabacteroides* 가 감소하였다. ($p<0.05$) 장내미생물의 변화와 대사증후군 관련지표 변화의 상관관계를 분석한 결과, 인슐린 저항성 지표인 HOMA-IR 이 Firmicutes/*Bacteroidetes* ratio 및 *Lactobacillus reuteri, Lactobacillus zeae, Clostridium butyricum, Roseburia sp* 의 존재비와 유의한 상관관계가 있었다. ($r>0.65$, $p<0.001$)

본 연구는 전통한식을 개량한 한국식 DASH 식단을 통해 한국인의 대사증후군 위험을 낮출 수 있는 가능성을 제시하였다. 전통한식의 취약점을 보완했기 때문에 한국식 DASH 식단은 서구형 식습관을 가진 군과 전통 한식 식습관을 가진 군에서 모두 대사증후군을 개선시키는 효과를 보였다. 또한 본 연구의 결과는 기존의 형성된 장내미생물총의 구성이 대사증후군관련 지표 개선에 관련이 있다는 가설을 뒷받침하고 있다. 보다 효과적인 대사증후군 관리를 위해서는 본 K-DASH 식단과 함께 전반적인 생활습관에 대한 행동 수정이 함께 이루어져야 할 것으로 보인다.

**주요어:** 대사증후군, 한국식 DASH 식단, 장내미생물, 인슐린 저항성, 한식의 건강효과, 메타지노믹스

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