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Does Exercise And Nutrition Style Affect Intestinal Microbiota Diversity?

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Summary. *Aim:* The aim of this study is to determine whether exercise and nutrition style has an effect on the intestinal microbiota diversity. *Materials and Method:* 15 healthy male volunteers between the ages of 18 and 24 participated in the study and they were grouped in three as professional athletes (n=5), amateur athletes (n=5) and sedentary individuals (n=5). "Stool samples" were taken from the participants once to find out their intestinal microbiota diversity, metagenomics analysis was conducted with New Generation Sequencing Method by using Illumina MiSeq analyzer and "Nutrient Consumption" questionnaire was given to find out their nutritional habits. Minitab 17 and SPSS 20.0 programs were used for the statistical analysis of the results and significance level was taken as $p<0.05$. *Results:* In our study which analyzed the species diversity, according to Shannon index, it was found that the group with the highest species diversity was the professional athletes group, while the group with the lowest species diversity was the sedentary individuals group. According to the Phylo diversity index, the group with the most phylo diversity was the professional athletes group, while the group with the least species diversity was the sedentary individuals group and a significant difference was found between amateur-professional, amateur-sedentary and professional-sedentary groups in terms of phylogenetic diversity ($p<0.05$). In addition, it was found that professional athletes had a diet consisting of protein predominantly, amateur athletes had a diet consisting of carbohydrates predominantly and sedentary individuals had a diet consisting of vegetable-fruit predominantly. *Conclusion:* it is thought that exercise influences intestinal microbiota diversity positively and different diets also have an influence in different microbial diversity between groups.

Key Words: Intestinal microbiota, Diversity, Exercise, Nutrition

Introduction

The gastrointestinal system is a complicated ecosystem that consists of hundreds of different microbial species and which hosts a highly developed microbial community. The human body includes approximately 10 trillion parenchyma cells; however, there are approximately 100 trillion microbial organisms within the community composition in the intestines depending on lifestyle and diet. These live bacteria which host a unique combination of about 500-1.000 different bacteria in every individual make up the intestinal microbiota in humans (1).

Intestinal microbiota, which generally lives in the mucosa of the digestive system, has various functions primarily in digestive and also in metabolic, physiological and immune system in the host organism. Intestinal microbiota, which begins to shape starting from the beginning of development, is shaped through being influenced by many factors such as diet, exercise, drug use, smoking and stress and microbial diversity has a significant effect on immune function, diseases and health (2).

The diversity and number of intestinal microbiota differ during a lifetime. The diversity of intestinal microbiota and the rates of strains are very important. With a wrong rate, some strains with a positive effect on health can turn into pathogens (3). The periods in which there is little diversity in intestines are considered as a risk factor in the development of diseases (4).

Along with sedentary life, food quality and low-value food consumption decrease the variety of intestinal flora and cause diseases (32). Loss or deterioration of intestinal microbiota diversity has been associated with many diseases such as autism, intestinal diseases, allergy, diabetes, cancer types and obesity associated with variable characteristics (5).

When the literature is reviewed, it can be seen that there are a great number of studies showing that exercise combined with a correct and healthy diet increases bacteria diversity in intestines and positively influences bacteria composition. Exercise prevents the occurrence of health problems, physical capacity, biochemical parameters, has a positive effect in reducing the level of stress. (6;33). It has also been found that exercise can increase the number of beneficial microbial species, enrich microflora diversity and heal the development of commensal bacteria (7). In the Gut Project, in a study conducted with 16 individuals, it was found that mild exercise had a positive influence on intestinal microbiota diversity (8). In a study analyzing the microbiota diversity of African and European children, it was found that European children had less intestinal microbiota diversity and more Firmicutes species, which is effective on obesity, since they have dietary programs in which there is more western ready food consumption and since they have immobile lifestyle (9). In a study that analyzed the intestinal microbiota of individuals who were given the Mediterranean diet, it was found that the number and diversity of healthy microbial species increased and also weight loss, lipid profile improvement and decrease in inflammation occurred in these individuals (10). It was found that extreme animal fat intake and too much sugar consumption caused a decrease in intestinal microbiota diversity and an increase in pathogen species (4).

In the regulation of number and diversity of useful species in intestinal microbiota, using factors such as changes in diet, pre/probiotic supplement and exercise can have an important place in preventing the formation of diseases by reversing the effects of intestinal germs on the body or in the treatment plan of diseases. In this context, this study is important as it will provide the response to the question that the way of exercise and nutrition will enable to have a healthy and diverse microbiota profile.

The aim of this study is to determine whether exercise and nutrition style has an effect on the intestinal microbiota diversity.

Materials and Methods

15 healthy male volunteers between the ages of 18 and 24 participated in the study. The participants were grouped in three as professional athletes (n=5), amateur athletes (n=5) and sedentary individuals (n=5). In the study, professional athlete group trained for 2 hours a day and 4 days a week, while the amateur group trained for 2 hours a day and 2 days a week. The sedentary group did not perform any physical activity.

In the study, criteria that could affect intestinal microbiota were determined beforehand and the participants who did not meet the criteria were excluded from the study. The inclusion criteria were as follows: not having used antibiotics at least for three months, not being on a diet, not using the probiotic and prebiotic supplement, not having any bowel disorder and in the athlete group having a sport age of 5 years and longer.

All the groups were informed about the procedure and the study a week before the study. Voluntary consent forms from participants and 40990478-050.99 dated and 20.06.2018 numbered ethical board approval report from Konya Selçuk University Faculty of Sport Sciences Deanship Non-interventional Clinical Researches Ethical Board were taken.

Nutritional and dietary habits of the participants were found by "Nutrient Consumption questionnaire" with face to face interview method. For microbiota analysis, stool samples of all participants were taken in 2 25 ml volume sterile spoon stool collection containers for one time only and kept at -20 C storage conditions. After all of the samples were collected, they were taken to the Medical Microbiology laboratory where analysis procedures would be performed.

DNA Isolation

200 mg of the stool sample taken from the participants was transferred to tubes with a glass bead of 0,11 mm diameter and 300 µL buffer (200 mM Tris-HCl, pH 8.0; 20 mM EDTA; 10% Triton X-100) and ho-

mogenized at 6000 rpm for one minute. 10 µl Lizozim (200 µg/µl) was added on the sample transferred to a new tube and incubated at 37 ˚C for 15 minutes. Later 250 µl lysis buffer (0,5 µg/µl Proteinase K, %5 Tween® 20, 3M Guanidinium thiocyanate, 20 mM Tris-HCl, pH 8.0) was added in the sample and incubated for 15 minutes at 70 ˚C and later for 5 minutes at 95 ˚C. Next, 250 µl isopropanol was added in the sample and was exposed to silica columns with centrifugation. DNAs bound to silica column were washed twice with wash buffer (20 mM NaCl, 2 mM Tris-HCl pH 7,5, %80 v/v Ethanol). DNA eluting was performed with 50 µL 100 mM Tris-HCl, pH 8,0 and kept at -20 ˚C until the DNAs were analyzed. The amount and quality of the DNA in samples DNA isolation of which were completed was measured with spectrophotometric methods and their suitability for subsequent steps was tested. Other molecular procedures were conducted with DNAs that had an OD260/OD280 rate of 1.8-2.0, OD260/OD230 rate of 2.0-2.2 and at least 10 ng/ul (preferably 50-300 ng/µL) concentration.

New Generation Sequencing (NGS)

Connector DNA sequences were added to the 5' end of target-specific primer pairs for compatibility with Illumina index and sequence adapters of the generated library. 16S rRNA-specific target primer-connector sequences were aligned as 5'TCGTCGGCAGCGTCAGATGTGTATAAGAGACAG-CCTACGGGNGGCWGCAG-3' for forward primer and as 5'-GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAG-GACTACHVGGGTATCTAATCC-3' for backward primer. The first PCR was applied by using "Biospeedy® Proof Reading DNA Polymerase 2x Reaction Mix" and 200 nm from each primer. The following thermal cycle program was followed in the PCR device: 3 minutes at 95°C; 25 cycles 30 seconds at 95°C, 30 seconds at 55°C and 30 seconds at 72°C; 5 minutes at 72°C. PCR product was purified by running an agarose gel, confirming the size (~550 bp) and by using "Biospeedy® PCR Product Purification Kit". The second PCR step and double index and Illumina sequencing adapters were added to purified first PCR sample by using Nextera XT Index Kit (Illumina, USA) and the following thermal cycle program was followed: 3 minutes at 95°C; 8 cycles 30 seconds at 95°C, 30 seconds at 55°C and 30 seconds at 72°C; 5 minutes at 72°C. PCR product was purified by using "Biospeedy® PCR Product Purification Kit" (Bioeksen, Turkey). The final library was confirmed for size (~630 bp) by using Bioanalyzer DNA 1000 chip. The final library was diluted to 4 nM by using 10 mM Tris pH 8.5 and aliquots of 5 µl were mixed to form a library pool. For clustering and sequencing preparation, the pooled libraries were denatured with NaOH, diluted with hybridization buffer (HT1) and denatured with temperature before MiSeq sequencing. Illumina MiSeq v3 reaction kits were used in runs. Minimum 5% PhiX was added in each reaction for internal control. Raw sequence data (the one with combined forward and backward reading) was sorted out by using the Qiime2 2018.11 version (https://qiime2.org/), degraded and analyzed. First index and primer sequences were trimmed and later original sequences were completed. The unaligned sequences at both ends of the sequences were removed by filtering and error check was performed. Contamination was prevented by pre-clustering. For Chimera elimination, settled UCHIME (11) code was used. Sequences were classified by using a settled classifier for Qiime2. Reference and taxonomy will be obtained from Qiime2 database. After the operational taxonomic unit (OTU) was chosen and the taxonomic assay was made according to the Qiime2 database, OTUs were grouped according to their phylotypes. The obtained microbial community profiles were compared with each other by using R software and dendrograms were formed. Emperor software was used to calculate PCA ordinations and the subsequent correlation analysis. $0.05 \geq p$ results were considered as statistically significant.

The following indices were used to find out the intestinal microbiota between groups.

Alpha Diversity (α-Diversity)

Alpha diversity (α-diversity) is the diversity in a single ecosystem or sample. The simplest criterion is the number of species (or OTUR) or richness observed in the sample. In order to calculate the diversity of species in a specific area or habitat, indices defined with specific functions are used. These indices are called diversity index. A diversity index is the mathematical measurement of species diversity in a population. Di-

Table 1. Age, height, weight values of groups

		n	x̄ ± sd	Minimum	Maximum
Age (years)	Professional footballer	5	18.00±0.00	18	18
	Amateur footballer	5	18.80±1.30	18	21
	Sedentary	5	21.80±1.30	21	24
Height (cm)	Professional footballer	5	181.20±3.96	176	187
	Amateur footballer	5	182.80±5.21	175	187
	Sedentary	5	174.20±2.38	170	176
Body weight (kg)	Professional footballer	5	74.00±5.78	68	83
	Amateur footballer	5	73.20±7.91	65	84
	Sedentary	5	68.60±10.31	55	82

versity indices simply provide more information than species richness (that is, the number of species) about the composition of the community; in addition, they also take into consideration the relative abundance of different species. Diversity indices provide important information about the rarity and cooperation of species in a community. The ability to measure diversity in this way is an important tool in terms of understanding the structure of the society.

a. Shannon Diversity Index (H)

Shannon diversity index (H) is an index used to characterize the diversity of species in a community. It is used to measure the entropy (ambiguity or content of information) in the sequence of the text. The more the index value is, the higher the diversity is.

b. Phylogenetic diversity

Phylogenetic diversity is the measure of biological diversity including phylogenetic differences between species. It corresponds to "Branching is part of a cladogram and minimum branching path is minimum" and it is defined and calculated as the total of all these branching. Phylogenetic indices are used in spatial analyses of biological diversity resulting from the presence of phylogenetic branching and the presence of tools to analyze these. The most commonly used phylogenetic index is Faith's Phylogenetic Diversity (12). PD is the phylogenetic analog of taxon richness and it is expressed as the number of tree units in a sample.

Statistical Analysis

The obtained microbial community profile was compared by using Minitab 17 software (Minitab, England) and dendrograms were formed. Minitab 17 software was also used for the calculation of PCA ordinations and the following correlation analyses. The obtained data were analyzed with SPSS 24 (Statistical Package for the Social Sciences) program and descriptive statistics were used in data analysis; significance level was taken as $p<0.05$.

Results

The results of the present study are presented in the following tables respectively.

According to Shannon index, the highest value and species diversity was found in professional athletes, while the least species diversity was found in the sedentary group.

According to phylodiversity index, the group with the most phylodiversity was the professional athletes, while the group with the least phylodiversity was the sedentary group. In addition, a significant difference was found between amateur-professional, amateur-sedentary and professional-sedentary groups in terms

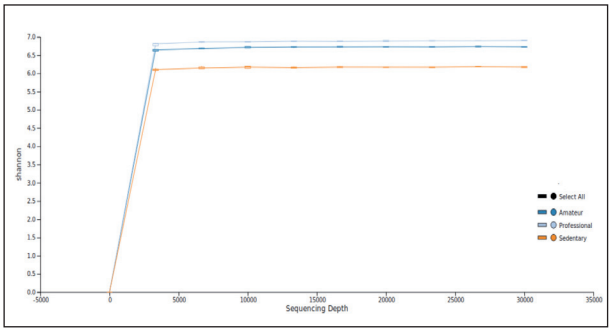

Figure 1. Shannon Diversity Index (H) of the groups

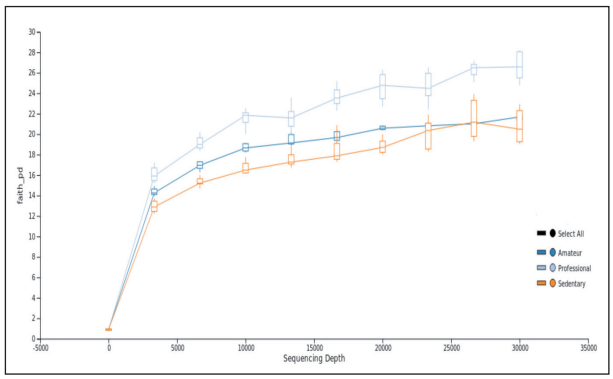

Figure 2. Phylogenetic diversity of the groups

of phylogenetic diversity ($p<0.05$).

It was found that the participants consumed meat, egg and legumes 3-5 times a week. It was found that 53% of professional athletes, 17% of amateur athletes and 30% of sedentary individuals consumed meat, egg and legumes 3-5 times a week. In the study, it was found that 37% of professional athletes, 14% of amateur athletes and 49% of sedentary individuals consumed fresh vegetable-fruit 1-2 times a week and 30% of professional athletes, 45% of amateur athletes and 2549% of sedentary individuals consumed bread and cereals 3-5 times a week.

Discussion

The results of our study which examined the microbial diversity and nutritional styles of professional and amateur athletes and sedentary individuals are discussed respectively below.

In our study, the groups with the highest species diversity according to Shannon and Phylogenetic diversity index values were professional athletes, amateur athletes and sedentary individuals, respectively.

In their study they examined the intestinal microbiota profile between active and sedentary women, Bressa et al., (2017) reported that low doses (max. 3 hours a week) of continuous physical activity modulated microbiota profile and increased the abundance of bacteria which support health in microbiota and they were associated with little microbiota richness in sedentary lifestyle and reported that further studies should be conducted to find out which doses and kinds of movement and exercise had an effect to increase microbiota diversity. In their study, Choi et al. (2013) found that there were differences between a total of 2510 bacteria taxon between rats which exercised and sedentary rats and that Lactobacillus arrangement was more in rats which exercised. Codella et al. (2017) found that as a homeostatic stimulant, exercise increased the number of benign microbial communities and diversified the intestinal microbiota. In another study on rats, Mika et al. (2016) found that exercise was influential on intestinal microbiota and BMI and exercise from early ages had positive effects on intestinal microbiota. In another study, it was reported that exercise had positive effects on the number and diversity of useful bacteria in intestinal microbiota (17). The rats which were randomly grouped in three were assigned to sedentary, voluntary wheel turning (low intensity) and compulsory treadmill exercise (high intensity) groups. It was found that microbial community structures of the three groups clustered separately in the intestinal region and unlike the hypothesis of the study group, voluntary wheel turning (low intensity) group had decreased bacteria richness when compared with the sedentary and obligatory treadmill exercise (high intensity) group. In addition, it was reported that bacterial taxons of both wheels turning and treadmill exercise rats changed. According to the results of this study, it was found that voluntary wheel and compulsory treadmill exercise changed the intestinal microbiome of the rats differently (18). In a study conducted with male rats, it was found that voluntary wheel turning exercise increased microbial diversity and caused increase especially in the genus of Actinobacteria and Lactobacillus (9). In a study which examined the effect of Tai-Chi exercise on intestinal microbiota, the existing evidence showed that exercise can increase diversity in intestinal microbiota and this diversity could be affected by diet, genetic and environmental factors and it was reported that optimal intensity, frequency and duration of exercise should also be known for useful effects on intestinal microbiota (19). In their study conducted on rats, Houghton et al. (2017) found that exercise did not have an effect on intestinal microbiota dysfunction; however, exercise increased intestinal microbiota diversity. In another study that compared the microbiota of rugby players, obese and healthy

Table 2. Distribution of nutrient consumptions of the samples according to groups

NUTRIENT		Frequency of consumption											
		Everyday		3-5 times a week		1-2 times a week		Every 15 days		Once a month		Never	
MEAT, EGG, LEGUMES													
Red meat	Professional	-	-	3	60	1	20	-	-	-	-	1	20
	Amateur	1	20	1	20	1	20	1	20	1	20	-	-
	Sedentary	-	-	2	40	1	20	1	20	1	20	-	-
Chicken	Professional	-	-	4	80	1	20	-	-	-	-	-	-
	Amateur	-	-	-	-	4	80	-	-	1	20	-	-
	Sedentary	-	-	3	60	2	40	-	-	-	-	-	-
Fish	Professional	-	-	-	-	5	100	-	-	-	-	-	-
	Amateur	-	-	-	-	2	40	2	40	-	-	1	20
	Sedentary	-	-	-	-	1	20	2	40	2	40	-	-
Egg	Professional	2	40	2	40	1	20	-	-	-	-	-	-
	Amateur	-	-	2	40	1	20	-	-	-	-	2	40
	Sedentary	4	80	1	20	-	-	-	-	-	-	-	-
Legumes (dry bean, chickpea, lentil)	Professional	-	-	4	80	1	20	-	-	-	-	-	-
	Amateur	-	-	1	20	3	60	-	-	1	20	-	-
	Sedentary	1	20	1	20	2	40	-	-	1	20	-	-
FRESH VEGETABLE-FRUIT													
Green-leafy vegetables	Professional	1	20	1	20	2	40	-	-	-	-	1	20
	Amateur	1	20	3	60	-	-	1	20	-	-	-	-
	Sedentary	2	40	1	20	1	20	-	-	1	20	-	-
Other vegetables	Professional	1	20	1	20	2	40	-	-	-	-	1	20
	Amateur	1	20	3	60	-	-	1	20	-	-	-	-
	Sedentary	-	-	-	-	4	80	-	-	1	20	-	-
Other fruit	Professional	1	20	3	60	1	20	-	-	-	-	-	-
	Amateur	-	-	1	20	3	60	-	-	-	-	1	20
	Sedentary	-	-	-	-	4	80	1	20	-	-	-	-
BREAD AND CEREALS													
White meat	Professional	3	60	1	20	-	-	-	-	-	-	1	20
	Amateur	3	60	2	40	-	-	-	-	-	-	-	-
	Sedentary	5	10	-	-	-	-	-	-	-	-	-	-
Wholemeal bread	Professional	1	20	2	40	-	-	1	20	-	-	1	20
	Amateur	-	-	-	-	1	20	-	-	1	20	3	60
	Sedentary	-	-	-	-	-	-	-	-	2	40	3	60
Rice	Professional	-	-	1	20	2	40	2	40	-	-	-	-
	Amateur	-	-	1	20	4	80	-	-	-	-	-	-
	Sedentary	-	-	1	20	4	80	-	-	-	-	-	-
Bulghur	Professional	-	-	2	40	1	20	1	20	-	-	1	20
	Amateur	-	-	1	20	4	80	-	-	-	-	-	-
	Sedentary	-	-	-	-	1	20	2	40	-	-	2	40
Pasta	Professional	-	-	3	60	2	40	-	-	-	-	-	-
	Amateur	-	-	1	20	4	80	-	-	-	-	-	-
	Sedentary	-	-	1	20	3	60	1	20	-	-	-	-
Bakery products	Professional	-	-	1	20	4	80	-	-	-	-	-	-
	Amateur	-	-	1	20	3	60	-	-	1	20	-	-
	Sedentary	1	20	-	-	-	-	3	60		-	1	20

subjects, it was found that rugby players had healthier species and diversity (21).

The factor of diet plays a key role in shaping microbial diversity. In our study, it was found that professional athletes had a protein dominated dietary style, while amateur athletes had a carbohydrate dominated dietary style and sedentary individuals had a vegetable-fruit dominated dietary style.

In a study in which a high-fat diet (with and without exercise) and normal diet (with and without exercise) were applied on rats, striking values were found in the microbiota profiles compared. It was found that exercise with both high fat and normal diet caused great changes in Bacteroidetes and Tenericutes phylum and a decrease in these species (22). In a study conducted with athletes, it was found that the athlete group consumed more protein when compared with the control group and they had a microorganism profile that had a high diversity representing 22 different phylum positively correlated with creatine kinase. In addition, it was reported that exercise played a significant role in the relationship between microbiota, host immunity and host metabolism; however, it was also reported that microbiota diversity is a complex issue including diet (5). Estaki et al. (2016) found that intestinal microbiota diversity was associated with aerobic fitness in healthy individuals and diet protein moderated microbial community composition. In another study, Genç et al. (2019) found that professional athletes who had predominantly protein diet had more clostridium species when compared with sedentary individuals. In one study conducted on rats, it was found that insufficient pomace intake with diet decreased microbial diversity and caused significant changes in microbiota composition (25). In intestinal microbiota of individuals who had Mediterranean diet, Prevotella, Lactobacillus and Bifidobacterium bacteria rates and fecal SCFA level were found to be high (26). In a study which examined the intestinal microbiota of women who lived in urban and rural areas, women who lived in the rural area were found to have more microbial diversity due to reasons such as less environmental pollution and natural diet (27). In a study conducted, it was reported that irrespective of diet, exercise increased microbial diversity; exercise capacity could be influenced by the presence of various microbiota and high-fat diet increased intestinal inflammation and exercise could decrease this inflammation and increase intestinal epithelium integrity (28). In a study by Wu et al. (2011), long term consumption of a diet rich in animal protein and saturated fat was associated with Bacteroides group (enterotype 1), while long term consumption of a diet rich in complex carbohydrate was associated with Prevotella group (enterotype 2). It was found that when compared with low-fat diets, high-fat diets significantly decreased fecal short-chain fat acid concentration and bifidobacterium count. In a study conducted on rats, the high fat diet was associated with low Lactobacillus intestinalis and species producing high amounts of propionate and acetate such as Clostridiales, Bacteroides, Enteroabacteriales. In addition, it was reported that in rats which consumed high-fat diet, microbial changes in the intestine influenced metabolic endotoxemia induced inflammation (30). Kopf et al. (2018) reported that a diet rich in vegetables could create an antiflammatory effect by changing microbial diversity.

In our study, it was found that professional and amateur exercise had a positive influence on intestinal microbiota diversity and that the results were in parallel with the literature. It is thought that the difference in preferred dietary styles has an effect on the different microbial diversity between groups. Conducting future studies that include a greater population and different branches and training methods and which examine different dietary styles of same level athlete groups will contribute to healthier results in this field.

References

1. Tannock GW. New perceptions of the gut microbiota: implications for future research. Gastroenterol Clin North A, 2005; 34(3):361–382.
2. Mika A, Fleshner M. Early-life exercise may promote lasting brain and metabolic health through gut bacterial metabolites. Immunology and Cell Biology, 2016; 94(2): 151.
3. Perlmutter D. Loberg K. Brain and intestine. İstanbul: Pegasus Publications 2017.
4. Mayer E. Brain-Intestine Connection. İstanbul: Paloma Publisher 2017.
5. Clarke SF, Murphy EF, O'Sullivan O et al. Exercise and associated dietary extremes impact on gut microbial diversity. Gut, 2014; 63:1913–1920.
6. Bilici MF, Güler M . The Investigation of the Acute Effect of Sparring Training on Some biochemical parameters

in Elite Boxers. International Journal of Applied Exercise Physiology, 2019; 8(3.1):357-360.
7. Monda V, Villaono I, Messina A et al. Exercise Modifies the Gut Microbiota with Positive Health Effects. Oxidative Medicine and Cellular Longevity, 2017;1-8.
8. Mach N, Fuster-Botella D. Endurance exercise and gut microbiota: A review. Journal of Sport and Health Science, 2017; 6(2):179-197.
9. Yatsunenko T, Rey FE, Manary MJ et al. Human gut microbiome viewed across age and geography. Nature, 2012; 486(7402): 222–227.
10. Singh A, Zapata RC, Pezeshki A, Reidelberger RD, Chelikani PK. Inulin fiber dose-dependently modulates energy balance, glucose tolerance, gut microbiota, hormones and diet preference in high-fat-fed male rats. J. Nutr. Biochem, 2018; 59:142–152.
11. Edgar Robert C et al. "UCHIME improves sensitivity and speed of chimera detection." Bioinformatics, 2011; 2194-2200.
12. DP Faith. Conservation evaluation and phylogenetic diversity. Biological Conservation, 1992; 61: 1-10.
13. Bressa C, Andrino AB, Santiago JP et al. Differences in gut microbiota profile between women with active lifestyle and sedentary women. PLOS ONE, 2017; 12(2):1-20.
14. Choie JJ, Eum SY, Rampersaud E, Daunert S, Abreu MT, Toborek M. Exercise attenuates PCB-induced changes in the mouse gut microbiome. Environ Health Perspect, 2013;121:725–730.
15. Codella R, Luzi L, Terruzzi I. Exercise has the guts: How physical activity may positively modulate gut microbiota in chronic and immune-based diseases. Digestive and Liver Disease, 2017;1-48.
16. Mika A, Fleshner M. Early-life exercise may promote lasting brain and metabolic health through gut bacterial metabolites. Immunology and cell biology, 2016;1-9.
17. Šket R, Treichel N, Kublik S et al . Hypoxia and inactivity related physiological changes precede or take place in absence of significant rearrangements in bacterial community structure: The PlanHab randomized trial pilot study. PLOS ONE, 2017; 12(12): 0188556.
18. Allen JM, Berg Miller, ME, Pence BD, Whitlock K, NehraV, Gaskins HR, White BA, Fryer JD, Woods JA.Voluntary and forced exercise differentially alters the gut microbiome in C57BL/6J mice. Journal of Applied Physiology, 2015; 118(8):1059-1066.
19. Hamasaki H. Exercise and gut microbiota: clinical implications for the feasibility of Tai Chi. Journal of Integrative Medicine, 2017; 15(4): 270-282.
20. Houghton D, Stewart DJ, Day DP, Trenell M. Gut Microbiota and Lifestyle Interventionsin NAFLD. International Journal of Molecular Sciences, 2016;17(447):2-29.
21. Barton W, Penney NC, Cronin O, Garcia-Perez I, Molloy MG, Holmes E, Shanahan F, Cotter PD O'Sullivan O. The microbiome of professional athletes differs from that of more sedentary subjects in composition and particularly at the functional metabolic level. Gut, 2018; 67(4):625-633.
22. Kang SS, Jeraldo PR, Kurti A et al. Diet and exercise orthogonally alter the gut microbiome and reveal independent associations with anxiety and cognition. Molecular Neuro degeneration, 2014; 9(1): 36.
23. Estaki M, Pither J, Baumeister P, Little JP, Gill SK, Ghosh S, Vand ZA, Marsden KT, Gibson, DL Cardiorespiratory fitness as a predictor of intestinal microbial diversity and distinct metagenomic functions. Microbiome, 2016;4(42): 2-13.
24. 24.Genç A, Tutkun E, Acar H, Zorba E. Investigation of Relation Between Clostridium Colonization and Nutrient Consumption in Intestinal Flora in Athletes and Sedentary Men. Progr Nutr [Internet]. 2019Apr.4 [cited 2019Sep.5]; 22(2).
25. Sonnenburg ED, Smits SA, Tikhonov M, Higginbottom SK, Wingreen NS, Sonnenburg JL. Diet-induced extinctions in the gut microbiota compound over generations. Nature, 2016, 529(7585): 212.
26. Fillippis FD, Pellegrini N, Vannini L et al. High-level adherence to a Mediterranean diet beneficially impacts the gut microbiota and associated metabolome. Gut, 2015; 65(11): 1812-1821.
27. Shin JH, Sim M, Lee JY, Shin DM. Lifestyle and geographic insights into the distinct gut microbiota in elderly women from two different geographic locations. Journal of Physiological Anthropology, 2016; 35(31): 2-9.
28. Campbell S, Wisniewski PJ. Exercises is a novel promoter of intestinal health and microbial diversity. Exercise and Sports Sciences, 2017;45(1): 41-47.
29. Wu J, Xu S, Xiang C, Cao Q, Li Q, Huang J, Shi L, Zhang J, Zhan Z .Tongue coating microbiota community and risk effect on gastric cancer. Journal of Cancer, 2018; 9(21): 4039-4048.
30. Cani PD, Possemiers S, Van de Wiele T et al. Changes in gut microbiota control inflammation in obese mice through a mechanism involving GLP-2-driven improvement of gut permeability. Gut, 2009;58: 1091–1103.
31. Kopf JC, Suhr MJ, Clarke J, Eyun SI, Riethoven JM, Ramer-Tait AE, Rose DJ. Role of whole grains versus fruits and vegetables in reducing subclinical inflammation and promoting gastrointestinal health in individuals affected by overweight and obesity: A randomized controlled trial. Nutr. J., 2018; 17:72.
32. Çağlayan Tunç A. Nutrition and obesity. İstanbul: Güven Plus Publisher 2019.
33. Çağlayan Tunç A. Examination of the relationship of personality structures of university students with time management and leisure time satisfaction. Gazi University Health Sciences Institute Doctoral thesis, Ankara.2019.

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