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피부 마이크로바이옴의 요인과 상호작용이 유해균에 미치는 영향에 대한 연구

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Influencing Factors and Interactions among the Skin Microbiomes in Affecting Detrimental Bacteria

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요 약: 본 연구는 우리나라 국민 20 ~ 49 세 연령군 가운데 289 명을 대상으로 피부 마이크로바이옴의 요인들인 유익균, 상재균, 여드름균이 유해균에 미치는 영향과 상호작용을 실증적으로 분석하기 위해 수행되었다. 피부 마이크로바이옴의 바이오 빅데이타를 활용하여 다중회귀모형으로 분석한 결과, 연구대상의 성별과 연령에 따른 피부 마이크로바이옴의 차이를 통제한 경우, 유익균은 유해균에 부(−)의 영향을 나타내었고, 상재균과 여드름균은 정(+)의 영향을 나타냈다. 특히 유익균이 유해균에 미치는 부(−)의 영향은 상재균의 수준에 따라 여드름균과의 상호작용을 통해 다르게 나타났다. 이러한 결과는 유익균의 활성화가 유해균을 억제하며, 피부 마이크로바이옴이 유해균에 미치는 영향은 독립적인 영향과 함께 상호작용을 통해서 피부 마이크로바이옴의 균형을 이루고 있음을 실증적으로 검증한 것이다. 따라서 개인 맞춤형 화장품 제조산업에서 유익균의 증식을 돕고 유해균을 억제하는 피부 환경을 조성하기 위한 피부 마이크로바이옴 제품을 연구할 때, 반드시 피부 마이크로바이옴 요인들의 독립적인 영향과 상호작용을 함께 고려해야 함을 시사한다.

Abstract: This study was conducted to empirically analyze the effects and interactions among beneficial bacteria, commensal bacteria, and acne bacteria, which are factors in the skin microbiomes, on detrimental bacteria by 289 people, who are 20 to 49 years old among Koreans. As a result of multiple regression models using bio big data of skin microbiomes, when the difference in skin microbiomes according to the sex and age of the subjects was controlled, the beneficial bacteria showed a negative (-) effect on the detrimental bacteria, while the commensal and acne bacteria showed a positive (+) effect. Particularly, the negative (-) effect of beneficial bacteria on detrimental bacteria was different through interaction with acne bacteria according to the level of commensal bacteria. These results demonstrate that the activation of beneficial

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bacteria inhibits detrimental bacteria, and the effect of skin microbiomes on detrimental bacteria is balanced with skin microbiomes through interaction with independent influence. Therefore, it is suggested that when studying skin microbiomes products to help the proliferation of beneficial bacteria and to create a skin environment that inhibits detrimental bacteria in the personalized cosmetics manufacturing industry, it is necessary to consider the independent effects and interactions among skin microbiome factors together.

Keywords: *skin microbiomes, bio big data, interaction effect, personalized cosmetics, multiple regression*

1. Introduction

As the future high added value of the microbiome is rapidly growing and massive research projects have been carried out in many areas including medicine, food, and cosmetic manufacturing industries, each country that fosters the bio-industry is aggressively investing in its physical and human capabilities in relevant areas. Since 2014 when the next generation sequencing (NGS) was developed, the microbiome technologies and patents began to drastically increase. Korea has recently observed remarkable achievements in this field; 14 cases of cancer and 82 other diseases using microbiome NGS were applied, a research institute with a global skin microbiome map was established, and the world's first anti-aging skin microbiome was successfully discovered [1-3].

However, the share of the patents the Korean entities applied is still negligible reaching merely 0.7 to 3.1% and the foreign countries have attempted to develop the microbiome technology led by global pharmaceutical companies. Meanwhile, small or medium-sized venture companies have been the driving force in Korea[4]. The domestic researches on skin microbiomes are mostly focused on treating or improving skin inflammation[5], and a representative case has been performed to treat acne and atopic dermatitis using beneficial bacteria, or to develop gene recombination strains using skin bacteria and to utilize useful proteins as delivering bodies[6]. In other words, domestic studies on the skin microbiomes are only at the level of revealing the characteristics of microbiome composition among normal people and skin diseases; or analyzing changes in microbiome composition before and after treatment to reveal that certain microbes have a preventive effect on dermatitis[7,8].

The microbiome is a combination of microbe and biome[9], which is the sum of the microbes that live in the body and means the whole of microbes and their genetic information [8,10]. 95% of microbiomes are distributed in the intestines, the digestive system, and also exist in the skin, respiratory tract, and oral cavity. Various microbiomes coexist on the skin in direct contact with the external environment, protecting detrimental pathogens from penetration into our body, responding to pathogens, and participating in local immune reactions[11]. Although the skin microbiomes exhibit a wide variety of distributions depending on internal factors such as gender, age, genetic characteristics, eating habits, and external factors such as geographic location, climate, ultraviolet exposure, and physical activity[12,13], it has been reported that in healthy adults the majority belong to *Actinobacteria* (52%), *Firmicutes* (24%), *Proteobacteria* (17%), and *Bacteriodetes* (7%), which have been identified as four phylum[14,15]. On the other hand, the skin microbiomes also vary greatly across races and countries[16,17], especially to understand the changes in the case of disease morbidity, and to carry out the therapeutic application and personalized cosmetic manufacturing design, basic data on the distribution and characteristics of the skin microbiomes of healthy people in each country is essential.

By the way, the results of basic research and analysis of the skin microbiomes of healthy people in Korea have been insufficient. The composition of microbiomes has been reported differently from different research conducted, and there is a limit that the general composition distribution of microbiomes among Koreans has not been officially established[7,18]. Moreover, most of the previous studies related to skin microbiomes are difficult to apply to generalize the results by conducting clinical trials with small samples [19,20]. Therefore, scientific research and empirical analysis using bio big data are essential to systematically develop the

cosmetics industry using the skin microbiomes, just as the characteristics and functions of intestinal microbiomes derived through many studies and analyses have been developed so far. While the previous version of 'customized cosmetics' is defined as the products classified based on the individual's skin condition and style, this 'up-to-date' version has become possible to implement customized cosmetics that meet the needs of consumers using information and communication technologies (ICTs) and biotechnologies (BTs)[6]. Nevertheless, there are few firms in this customized cosmetics market that provide customized cosmetics based on microbiomes of different skin for each individual.

This study aimed to analyze the composition of microbiomes distributed on facial skins of ordinary people in Korea aged the 20 to 40s, who had been recognized that there were no specific skin troubles, and to empirically study the functional aspects of microbiomes to provide and support the information and data necessary for the personalized cosmetics manufacturing industry. In particular, the gender and age that had the greatest influence on the characteristics of the skin microbiomes were controlled, and the effects of beneficial bacteria, commensal bacteria, and acne bacteria, which were a general composition of the skin microbiomes, on detrimental bacteria were analyzed.

Furthermore, it was analyzed whether the interaction exists empirically among beneficial bacteria, commensal bacteria, and acne bacteria that affect detrimental bacteria. Each microbial group existing in the human body does not independently affect the intestines and skin, but forms a balance of microbiomes as an ecosystem that interacts with each other[21]. Especially, skin microbiomes, which are micro-organisms such as bacteria, fungi, and viruses, exist on the skin that is directly exposed to the external environment, and the balance of the skin microbiomes appears differently when the composition of a specific microbiome is changed[22,23]. The imbalance of the skin microbiomes can also cause inflammation such as atopic dermatitis. And it has been reported that the *Staphylococcus aureus* (*S. aureus*) is less than 5% of the skin in normal individuals, compared to over 90% among most atopic dermatitis patients[24]. In addition, the increase of *S. aureus* decreased the diversity of skin bacteria and this was also found to be correlated with the prevalence of atopic dermatitis[24]. These previous studies commonly derived that *S. aureus* is a very important microorganism for atopic dermatitis and interacts with other microorganisms.

Personalized cosmetics use the interaction of the skin microbiomes to help create a skin ecosystem environment so that the balance of broken skin microorganisms is close to the skin microbiomes composition of healthy normal people. In particular, the main goal of the personalized cosmetics manufacturing industry is the development of ingredients that increase the growth of beneficial bacteria that are beneficial to skin health while suppressing detrimental bacteria that are detrimental to skin health among the microbes that make up skin microbiomes[5,19]. For this purpose, the interaction of the skin microbiomes based on empirical data will be verified and the findings should be derived from the clinical trials.

2. Materials and Methods

2.1. Research Hypothesis and Model

The purpose of this study is to provide the basic data and information to the customized cosmetics manufacturing industry by confirming whether interactions among the skin microbiomes appear through empirical analysis of the skin microbiomes affecting detrimental bacteria. To this end, three research questions and research model were set up as follows.

Research Question 1: The composition ratio of skin microbiomes will be different according to the internal factors of skin microbiomes, gender and age group.

Research Question 2: When gender and age are controlled as internal factors affecting skin microbiomes, beneficial bacteria will have a negative (-) effect on detrimental bacteria, and commensal bacteria and acne bacteria will have a positive (+) effect.

Research Question 3: There will be interactions among the skin microbiomes ecology that affects detrimental bacteria.

Figure 1 shows a research model that schematizes the research hypothesis set out above.

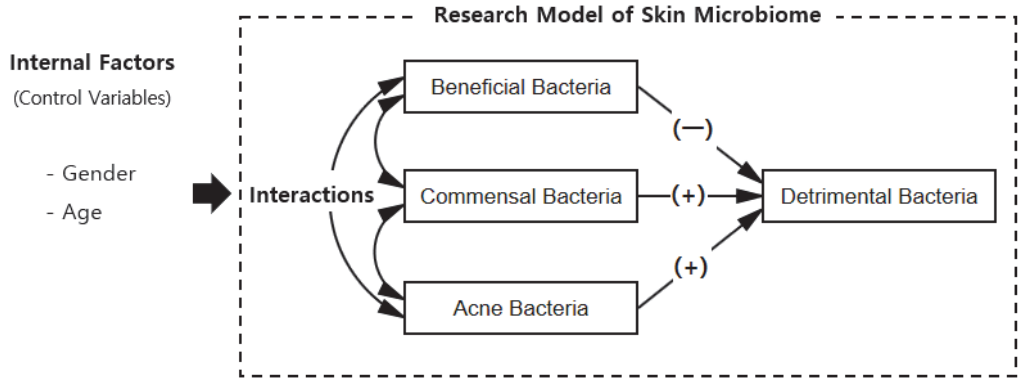


Figure 1. Research model: this is a multiple regression model. Among the internal factors of skin microbiome, gender and age were designed as control variables, and the explanatory variables, beneficial bacteria, commensal bacteria, and acne bacteria, were visualized as independent effects and their interactions on detrimental bacteria as dependent variable.

2.2. Study Participants and Data Collection

To verify the questions of this research, the skin microbiomes were collected from adult males and females aged 20 to 49 years across the country. The subjects were excluded from the study for cases of infectious skin disease[25] or trauma such as abrasions at the sampling site[26]. Skin samples were collected by swabbing the forehead (a 4 × 2 cm area of the center) using a sterile swab kit (Shebah Biotech Inc., Korea) after removing their makeup. Metagenomic deoxyribonucleic acid (DNA) was extracted from the skin samples using the Nucleospin® Tissue XS kit (MACHEREY-NAGEL, Germany). Polymerase chain reaction (PCR) amplification of microorganism-derived genomic DNA was conducted using a TProfessional Trio Thermocycler (Biometra, Germany). The PCR products were fractionated by electrophoresis in 2% (w/v) agarose gels (Sigma-Aldrich, USA), stained with RedSafe (Intron Biotechnology Inc., Korea) and visualized under the Gel Doc XR+ imaging system (Bio-Rad, USA). Subsequently, density of bands derived from PCR products were measured using the Gel Doc XR+ imaging system and quantification of the density was conducted using an Image Lab software (Ver 6.0, Bio-Rad, USA). All primers were designed by Primer3 software (Whitehead Institute/MIT Center for Genome Research, USA) with variable region of 16S ribosomal RNA derived from each target microorganism. In this process, bio big data of the skin microbiomes were collected using the Image Lab program. Among the collected data, 26 genus of the top 1% with the largest total amount were selected as analysis data, and a total of 7,514 skin microbiomes big data were secured by synthesizing 26 genus of 289 subjects.

2.3. Study Tools

2.3.1. Sociodemographic Factors

As demographic variables, gender and age, which were the main factors of the skin microbiomes, were designed. Gender was measured for males and females, and age was measured for 289 people in their 20s and 40s. Skin microbiomes were distributed differently according to gender[27], and children, teenagers, and age groups after 50 have various body changes due to growth and aging[28], so they were excluded from this study.

2.3.2. Skin Microbiomes

Skin microbiomes in which micro-organisms live in clusters on the skin are classified as beneficial bacteria[29], commensal bacteria[30], and detrimental bacteria[31]. Beneficial bacteria have the function of activating immunity and preventing diseases and inflammation caused by the skin, and detrimental bacteria cause skin problems and diseases while causing a decrease in immunity. Commensal bacteria, known as intermediate bacteria, have the characteristic of attaching to and distributing to the predominant bacteria among beneficial or detrimental bacteria depending on the skin environment. In

Table 1. Skin Microbiome Group according to the Characteristics of the Genus

No.	Genus	Skin Microbiome Group	No.	Genus	Skin Microbiome Group
1	*Lactobacillus rhamnosus HK-9*	Beneficial Bacteria[29]	15	*Staphylococcus epidermidis*	Commensal Bacteria[30]
2	*Lactobacillus paracasei HS-05*		16	*Psuedomonas koreensis*	
3	*Bacillus licheniformis*		17	*Propionibacterium qranulosum*	
4	*Bacillus subtilis*		18	*Streptococcus salivarius*	
5	*Staphylococcus aureus*	Detrimental Bacteria[31]	19	*Neisseria subflava*	
6	*Pseudomonas aeruginosa*		20	*Staphylococcus warneri*	
7	*Corynebacterium jeikeium*		21	*Pseudomonas tolaasii*	
8	*Corynebacterium diphtheriae*		22	*Acidovorax temperans*	
9	*Streptococcus pyogenes*		23	*Dietzia maris*	
10	*Streptococcus cristatus*		24	*Bradyrhizobium japonicum*	
11	*Acinetobacter junii*		25	*Acinetobacter calcoaceticus*	
12	*Finegoldia magna*		26	*Propionibacterium acne*	Acne Bacteria[32]
13	*Stenotrophomonas maltophilia*				
14	*Corynebacterium pseudogenitalium*				

this study, skin microbiome data were secured by dividing the skin microbiomes into beneficial bacteria, commensal bacteria, and detrimental bacteria according to the general classification. In particular, among detrimental bacteria, acne bacteria have a relatively large distribution and are the main cause of skin trouble[32], so it was designed together as a research variable.

As shown in Table 1, a total of 26 genus, which was relatively most distributed in the skin microbiomes, were classified into skin microbiome groups with similar characteristics such as beneficial bacteria, commensal bacteria, and detrimental bacteria. The beneficial bacteria were composed of two species, *Lactobacillus and Bacillus*, of which *Bacillus licheniformis* was more than 80%. The detrimental bacteria were composed of 9 species including *S. aureus*, and of which *Acinetobacter juni* and *Finegoldia magna* were relatively high. The *S. aureus* was composed of 10 species including *Staphylococcus epidermidis,* and of which *Pseudomonas tolaasii* and *Dietzia maris* were relatively high. And acne bacteria were set to *Propionibacterium acnes* single bacteria.

2.4. Data Analysis

The summary of the sociodemographic characteristics was presented by frequency analysis and the difference in the ratio of skin microbiomes by age was analyzed by the Chi-square independence test. To figure out the average difference in the skin microbiomes by gender, independent sample *t* test was performed. For the average difference test of skin microbiomes by age groups, one-way analysis of variance (ANOVA) was used, and then the Scheffe method was applied as a post hoc comparison test. In addition, correlation analysis was conducted to identify the linearity of detrimental bacteria and other skin microbiomes, and multiple regression analysis was conducted to study the characteristics of skin microbiomes affecting detrimental bacteria.

3. Results

3.1. Difference in the Ratio of the Skin Microbiomes by Gender and Age

The demographic characteristics of the individuals in this study were 77 males (26.6%) and 212 females (73.4%) by gender. As for the age of males, 26 people aged 20 years (20 ~ 29 years), 24 people aged 30 years (30 ~ 39 years), and 27 people aged 40 years (40 ~ 49 years). For the age of females, 59 people aged 20 years, 70 people aged 30 years, and 83 people aged 40 years.

Table 2 showed the ratio of skin microbiomes by gender

Table 2. Differences in the Proportion of the Skin Microbiomes by Gender and Age (N = 289, unit: ηg)

Age	Male	Beneficial Bacteria	Commensal Bacteria	Acne Bacteria	Detrimental Bacteria	*p*-value
20 ~ 29	N = 26	1,233 (5.2%)	12,271 (52.1%)	6,226 (26.4%)	3,811 (16.2%)	
30 ~ 39	N = 24	2,023 (8.2%)	11,023 (44.8%)	6,530 (26.5%)	5,043 (20.5%)	< 0.001
40 ~ 49	N = 27	1,259 (4.3%)	14,015 (47.6%)	6,428 (21.8%)	7,739 (26.3%)	
Total	N = 77	4,515 (5.9%)	37,309 (48.1%)	19,184 (24.9%)	16,593 (21.0%)	
Age	Female					
20 ~ 29	N = 59	3,198 (5.4%)	27,922 (47.1%)	15,247 (25.7%)	12,908 (21.8%)	
30 ~ 39	N = 70	4,132 (4.8%)	33,843 (39.6%)	17,161 (20.1%)	30,342 (35.5%)	< 0.001
40 ~ 49	N = 83	3,549 (4.1%)	39,916 (46.1%)	17,067 (19.7%)	26,066 (30.1%)	
Total	N = 212	10,879 (4.8%)	101,681 (44.3%)	49,475 (21.8%)	69,316 (29.1%)	

The *p*-values were obtained using the chi-square independence test.

and age, and there was a significant statistical difference. Males were distributed in the order of commensal bacteria (48.1%) > acne bacteria (24.9%) > detrimental bacteria (21.0%) > beneficial bacteria (5.9%), and females were significant in the order of commensal bacteria (44.3%) > detrimental bacteria (29.1%) > acne bacteria (21.8%) > beneficial bacteria (4.8%). The distribution of the skin microbiomes in male group was similar in their 20s and 30s, but in their 40s, detrimental bacteria were 26.3% and acne bacteria were 21.8%. Females' skin microbiomes distribution was similar in their 30s and 40s, but in their 20s, acne bacteria were 25.7% and detrimental bacteria were 21.8%.

3.2. Mean Difference Analysis of the Skin Microbiomes by Gender and Age

The descriptive statistics for beneficial bacteria, commensal bacteria, acne bacteria, and detrimental bacteria, which were skin microbiomes strains of the study data, were presented in Table 3. The average of beneficial bacteria was 187.30 ηg (nanograms) and the standard error was 9.29 ηg. The average of commensal bacteria was 48.84 ηg and the standard error was 4.36 ηg. The average of acne bacteria was 424.68 ηg and the standard error was 23.00 ηg, the average number of detrimental bacteria was 253.09 ηg and the standard error was 20.13 ηg.

Table 4 was a result of analyzing the average of the skin microbiomes by gender and age of the study data, and there was statistically no significant difference in average in beneficial bacteria, commensal bacteria, and acne bacteria, while there was a statistically significant difference in both gender and age groups only for detrimental bacteria.

In relation to the average of skin microbiomes for gender, the average of males for beneficial bacteria was 51.93 ηg, which tended to be higher than the average of 44.80 ηg for females. The average of commensal bacteria was 369.47 ηg for men, lower than 401.46 ηg for females. The average of acne bacteria was 173.78 ηg for males and 177.38 ηg for females, which was similar. The detrimental bacteria averaged

Table 3. Descriptive Statistics of the Skin Microbiomes (N = 289, unit: ηg)

	Descriptive statistics					95% Confidence Interval of Average	
	Min	Max	Median	Average	Standard Error	Lower Limit	Upper Limit
Beneficial Bacteria	0	624	162.50	187.30	9.29	169.02	205.59
Commensal Bacteria	0	3,664	19.80	48.84	4.36	40.26	57.42
Acne Bacteria	0	1,647	337.50	424.68	23.00	379.41	469.95
Detrimental Bacteria	0	4,082	167.70	253.09	20.13	213.46	292.72

Table 4. Mean Differences of the Skin Microbiomes by Gender and Age (n=289, unit: ηg)

Gender		Beneficial Bacteria	Commensal Bacteria	Acne Bacteria	Detrimental Bacteria
Male	Mean (SE)	51.93[a] (7.68)	369.47[a] (32.46)	173.78[a] (11.77)	174.84[a] (20.02)
Female	Mean (SE)	44.80[a] (4.34)	401.46[a] (18.80)	177.38[a] (7.80)	241.64[b] (15.12)
p-value		0.406	0.387	0.808	0.009
Age					
20 ~ 29	Mean (SE)	50.64[a] (6.75)	354.25[a] (24.15)	162.62[a] (11.50)	176.16[a] (17.52)
30 ~ 39	Mean (SE)	52.54[a] (8.01)	404.49[a] (28.73)	191.03[a] (10.96)	230.84[ab] (19.17)
40 ~ 49	Mean (SE)	38.56[a] (4.93)	414.08[a] (29.74)	174.96[a] (11.22)	256.26[b] (24.55)
p-value		0.242	0.288	0.225	0.028

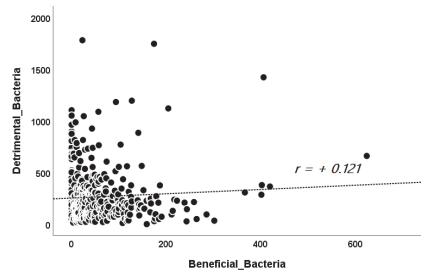
The *p*-values were obtained using the independent sample *t* test and one-way ANOVA.
As a result of Scheffe's multiple comparison analysis, if the superscript of the mean is the same, it means that there is no statistically significant difference. The SE means standard error.

174.84 ηg in males and lower than the average of 241.64 η g in females. But, as a result of performing the mean of skin microbiomes for gender with an independent sample *t* test, only detrimental bacteria had a statistically significant mean difference.

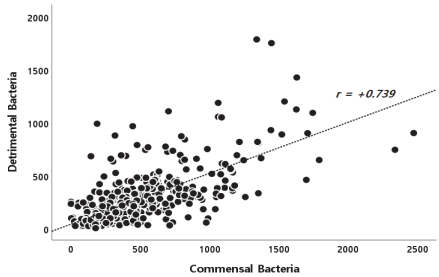
Regarding the skin microbiomes by age groups, beneficial bacteria had not changed to 50.64 ηg for 20s and 52.54 ηg in their 20s, but they tended to decrease rapidly to 38.56 ηg in their 40s. The average of commensal bacteria was 354.25 ηg in the 20s, and it increased rapidly to 404.49 ηg in the 30s, and tended to maintain to 414.08 ηg in the 40s. The average of acne bacteria was 162.62 ηg in the 20s, increased to 191.03 ηg in the 30s, but slightly decreased to 174.96 η g in the 40s. The average of detrimental bacteria was 176.16 ηg in the 20s, 230.84 ηg in the 30s and 256.26 ηg in the 40s, which tended to increase as the age increased. However, as a result of performing the statistical average difference on skin microbiome by age groups with ANOVA and performing multiple comparison on average with Scheffe, there was no statistically significant difference in beneficial bacteria, commensal bacteria, and acne bacteria by age groups, while there was statistically significant difference only in detrimental bacteria.

3.3. Correlation Analysis of the Skin Microbiomes by Gender

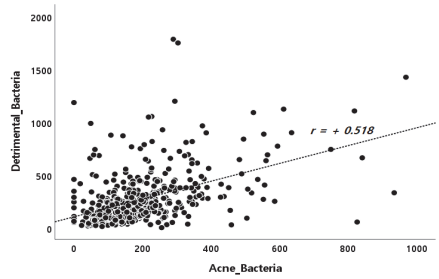
Figure 2 presented the scattering of detrimental bacteria and other skin microbiomes, and all correlation coefficients were



(A) Beneficial Bacteria and Detrimental Bacteria



(B) Commensal Bacteria and Detrimental Bacteria



(C) Acne Bacteria and Detrimental Bacteria

Figure 2. The correlation among the skin microbiomes: (A) beneficial bacteria and detrimental bacteria; (B) commensal bacteria and detrimental bacteria; (C) acne bacteria and detrimental bacteria.

Table 5. Correlation between Skin Microbiomes by Gender (N = 289)

Female \ Male	Beneficial bacteria	Commensal bacteria	Acne bacteria	Detrimental bacteria
Beneficial bacteria	1	+ 0.109	+ 0.354**	+ 0.137
Commensal bacteria	+ 0.171*	1	+ 0.724***	+ 0.755***
Acne bacteria	+ 0.222**	+ 0.792***	1	+ 0.689***
Detrimental bacteria	+ 0.055	+ 0.862***	+ 0.705***	1

The p-values were obtained using correlation analysis. $^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$

significant. The correlation coefficient of beneficial bacteria and detrimental bacteria showed a very weak positive correlation between + 0.121, while commensal bacteria and detrimental bacteria had a strong positive correlation of + 0.739. In addition, the correlation coefficient between acne bacteria and detrimental bacteria was + 0.518, which was a positive (+) correlation.

Table 5 was the result of analyzing the correlation between the skin microbiomes by gender. Based on the diagonal self-correlation 1, the right was male and the left was female. Both males and females did not correlate beneficial bacteria and detrimental bacteria, and the correlation coefficient of commensal bacteria and detrimental bacteria had a strong correlation between + 0.755 in males and + 0.862 in females. The correlation coefficient between acne bacteria and detrimental bacteria was + 0.689 for males and + 0.705 for females, indicating a significant strong positive correlation. And the correlation between commensal bacteria and acne bacteria had a strong correlation of + 0.724 for males, + 0.792 for females, beneficial bacteria and acne bacteria showed a positive weak correlation with + 0.354 in males and + 0.222 in females. Also, the correlation between beneficial bacteria and commensal bacteria was not significant in males, but there was a positive weak correlation in females with + 0.171.

3.4. Interactions among the Skin Microbiomes Affecting Detrimental Bacteria

Table 6 was a result of a multiple regression analysis of the characteristics of skin microbiomes that affect detrimental bacteria.

In the skin microbiomes, there was no difference in the

Table 6. Interactions among the Skin Microbiomes Affecting Detrimental Bacteria (N = 289)

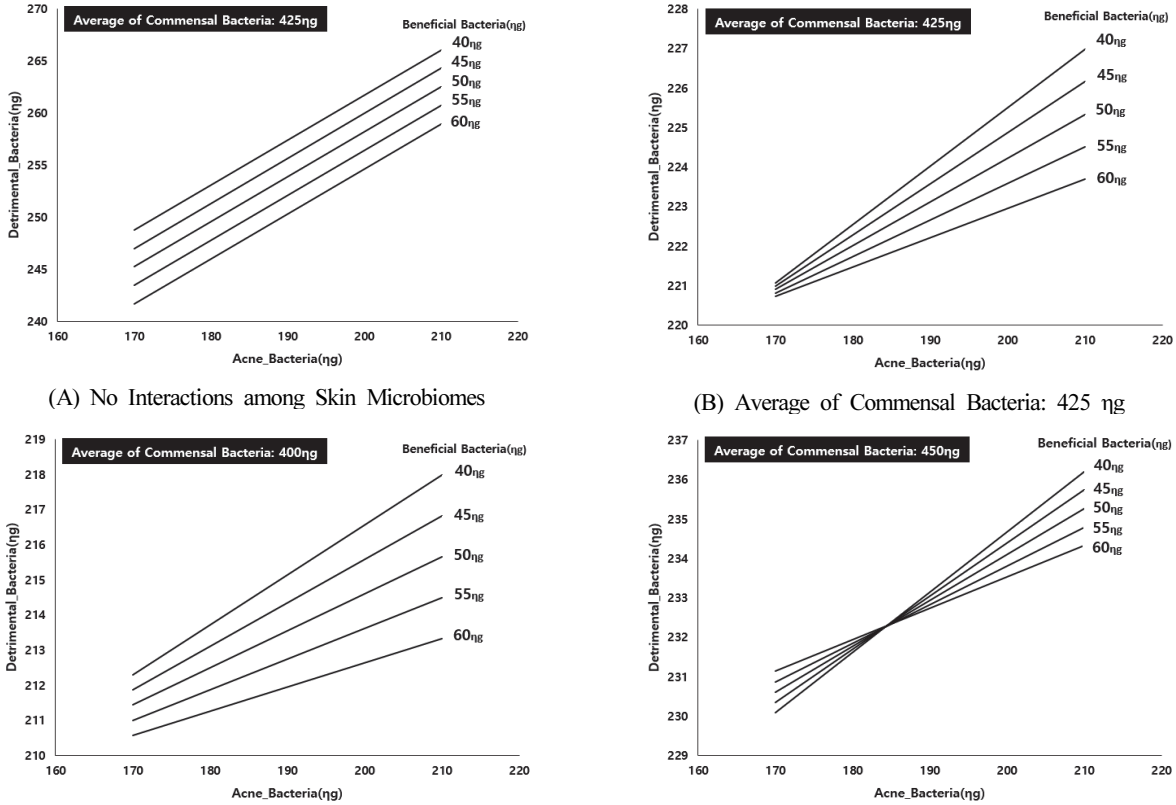
Independent Variables	Model 1			Model 2			
	β	Partial R	p-value	β	Partial R	p-value	VIF
(Constant)	284.794		< 0.001	- 61.819		0.238	
Gender	73.433	0.166	0.005	46.579	0.148	0.013	1.016
Age	2.824	0.121	0.041	1.798	0.108	0.070	1.030
Beneficial Bacteria	- 0.354	- 0.129	0.029	- 0.573	- 0.172	0.004	1.629
Commensal Bacteria	0.579	0.598	< 0.001	0.208	0.274	< 0.001	2.758
Acne Bacteria	0.431	0.215	< 0.001	0.203	0.111	0.043	3.311
Beneficial Bacteria × Commensal Bacteria				0.003	0.286	< 0.001	8.967
Acne Bacteria × Commensal Bacteria		-		0.001	0.682	< 0.001	2.089
Beneficial Bacteria × Acne Bacteria				- 0.004	- 0.228	< 0.001	9.145
Adjusted R^2	67.7%			83.6%			
$\triangle F$	-			92.669 (p-value < 0.001)			

The p-values were obtained using multiple regression analysis.

average of beneficial bacteria, commensal bacteria, and acne bacteria in the gender and age group, and only detrimental bacteria were different in the 20s and 40s. The characteristics of gender and age were designed as control variables as in Model 1, and the effects of independent variables such as beneficial bacteria, commensal bacteria, and acne bacteria on detrimental bacteria were analyzed. As a result, beneficial bacteria had a negative effect with a non-standardization coefficient of - 0.354, and commensal bacteria and acne bacteria had a positive effect with + 0.579 and + 0.431. The explanatory power of Model 1 was high at 67.7% and the variable that had relatively the most influence on detrimental bacteria was commensal bacteria with a partial correlation coefficient of 0.598.

Model 2 was designed by multiplying each variable in Model 1 to empirically analyze the interaction between beneficial bacteria and commensal bacteria, acne bacteria and commensal bacteria, and beneficial bacteria and acne bacteria, respectively. As a result of the analysis, the explanatory power of Model 2 went up high at 83.6%, which was significantly increased by 15.9%p from Model 1, and all variables designed by interaction terms showed statistically significant results. This result meant that all terms designed by interaction are significant, and it was confirmed that there were interactions among beneficial bacteria, commensal bacteria, and acne bacteria that affected detrimental bacteria. And variance inflation factor (VIF) was all less than 10, and there was no multicollinearity. In addition, the independent variable that had the most influence on detrimental bacteria was the interaction term between acne bacteria and commensal bacteria with the largest absolute value of the partial correlation coefficient of + 0.682, followed by the interaction term between beneficial bacteria and commensal bacteria.

Figure 3 was a graph using two multiple regression models



(A) No Interactions among Skin Microbiomes

(B) Average of Commensal Bacteria: 425 ηg

(C) - 1 Standard Error of Commensal Bacteria Average: 400 ηg

(D) + 1 Standard Error of Commensal Bacteria Average: 450 ηg

Figure 3. The interactions of the skin microbiomes: (A) No interactions among skin microbiomes; (B) Average (425 ηg) of commensal bacteria; (C) - 1 standard error (400 ηg) of commensal bacteria average; (D) + 1 standard error (450 ηg) of commensal bacteria average.

(i.e., Model 1 and Model 2) in Table 6 that analyzed the effects of the skin microbiomes that controlled for gender and age on detrimental bacteria.

Firstly, part (A) was a visualization of a model in which there was no interaction of the skin microbiomes, such as Model 1. The average 425 ηg of the commensal bacteria were fixed and acne bacteria were changed from 170 ηg, which was 95% lower limit of the average, to the upper limit of 210 ηg. And when the level of beneficial bacteria varied from 40 ηg, which was the lower limit of 95% of the average, to the upper limit of 60 ηg, the average of detrimental bacteria was calculated through Model 1 shown in a graph. Since there was no interaction among beneficial bacteria, commensal bacteria, and acne bacteria, the slope of the effect on detrimental bacteria was the same depending on the average level of beneficial bacteria when acne bacteria increased. In other words, even if the average of beneficial bacteria was low such as 40 ηg, or relatively high such as 60 ηg, the increase in the effect on detrimental bacteria were all calculated equally, and it indicated that beneficial bacteria, commensal bacteria, and acne bacteria had only independent effects on detrimental bacteria.

On the other hand, part (B) was a graphically visualized multiple regression model that took into account the interaction of the skin microbiomes analyzed in Model 2 of Table 6. In the same way as part (A), commensal bacteria were fixed at an average of 425 ηg, and when acne bacteria changed from the lower limit of 95% average to the upper limit of the average, the average of detrimental bacteria was calculated according to the level of acne bacteria. As a result, when the level of beneficial bacteria was low such as 40 ηg, the average slope of detrimental bacteria was large when the level of beneficial bacteria increased, and when the level of beneficial bacteria was high such as 60 ηg, the slope of detrimental bacteria was relatively low. Thus, it was confirmed that beneficial bacteria and acne bacteria had the effect of controlling the effects of harmful bacteria through interaction.

And the parts (C) and (D), after respectively fixing 400 η g calculated with - 1 standard error and 450 ηg calculated with + 1 standard error on the average of the commensal bacteria, were visualized as the average of detrimental bacteria calculated according to the level of beneficial bacteria when the acne bacteria changed according to 95% confidence interval such as (B). Part (C), which had a fixed level of 400 ηg of the commensal bacteria, showed similar characteristics to (B) when the level of beneficial bacteria was different depending on the fluctuations of acne bacteria. However, in part (D), where the average of the phase bacteria was fixed at 450 ηg, when the average of the acne bacteria changed and the level of the beneficial bacteria changed, the average of the detrimental bacteria was completely different from that of (B).

These results have shown empirically that when the level of the commensal bacteria varies from the same as the average, lower than average, and higher than average, since the interaction between acne bacteria and beneficial bacteria also shows different effects of detrimental bacteria, beneficial bacteria, commensal bacteria, and acne bacteria interact with

Table 7. Interactions among the Skin Microbiomes and Average of Detrimental Bacteria (N = 289, unit: ηg)

Average of Detrimental Bacteria	(A) No interactions			(B) Commensal Bacteria: 425 ηg			(C) Commensal Bacteria: 400 ηg			(D) Commensal Bacteria: 450 ηg		
	95% Confidence Interval (lower limit and upper limit) of Acne Bacteria Average (ηg)											
Beneficial Bacteria	170	210	△ηg	170	210	△ηg	170	210	△ηg	170	210	△ηg
40	248.8	266.0	+17.25	221.1	227.0	+5.90	212.3	218.0	+5.69	230.1	236.2	+6.12
45	247.0	264.2	+17.25	221.0	226.1	+5.17	211.9	216.8	+4.96	230.3	235.7	+5.39
50	245.2	262.5	+17.25	220.9	225.3	+4.44	211.4	215.7	+4.23	230.6	235.3	+4.65
55	243.4	260.7	+17.25	220.8	224.5	+3.70	211.0	214.5	+3.49	230.9	234.8	+3.92
60	241.7	258.9	+17.25	220.7	223.7	+2.97	210.6	213.3	+2.76	231.1	234.3	+3.19
△ηg	-7.084	-7.084		-0.354	-3.284		-1.723	-4.653		+1.051	-1.879	

each other to control their effects on detrimental bacteria.

Table 7 was the result of calculating the average of detrimental bacteria by applying the multiple regression model of the effect of skin microbiomes on detrimental bacteria for the four cases in Figure 3.

Part (A) was the case where there was no skin microbiome interaction, and (B), (C), (D) were calculated the average of detrimental bacteria when there were interactions in the skin microbiomes. That was, the average of commensal bacteria was fixed at 425 ηg, 400 ηg, and 450 ηg in the order of (B), (C), and (D), respectively. And if acne bacteria were changed from the 95% lower limit of 170 ηg to the upper limit of 210 ηg, how the average increases of harmful bacteria were calculated when the average of beneficial bacteria increased from the 95% lower limit of 40 ηg to the upper limit 60 ηg were analyzed and visualized.

The results of (A) showed that the interaction of skin microbiomes did not exist, so the increases in harmful bacteria were all the same. The average increase of detrimental bacteria was + 17.25 when the average of beneficial bacteria was fixed at 425 ηg, and when the average of acne bacteria was increased from 170 ηg to 210 ηg. And when the average of acne bacteria was 170 ηg, if the average of beneficial bacteria was increased from 40 ηg to 60 ηg, the average increase of detrimental bacteria was - 7.084.

Part (B) was the same as the design of (A) and confirmed the variation in the average of detrimental bacteria for the model in which additional interactions existed. As a result, if the average of beneficial bacteria was fixed at 40 ηg and the average of acne bacteria increased from 170 ηg to 210 ηg, the average increase of detrimental bacteria was + 5.90. And if the average of beneficial bacteria was fixed at 60 ηg, the increase of the average of detrimental bacteria was + 2.97. In other words, if beneficial bacteria were increased by 1.5 times, detrimental bacteria were reduced by about twice. On the other hand, if the average of acne bacteria was fixed at 170 ηg and the average of beneficial bacteria was changed from 40 ηg to 60 ηg, the increase in the average of detrimental bacteria was - 0.354. If the average of acne bacteria was fixed at 210 ηg, the increase in the average of detrimental bacteria was − 3.284.

In conclusion, it was confirmed that when the average of acne bacteria was about 1.2 times higher at 210 ηg than when the average of acne bacteria was 170 ηg and the average of beneficial bacteria was increased by 1.5 times, the average of harmful bacteria was decreased by about 9.3 times.

In the case of (C), the average of the commensal bacteria was fixed at 400 ηg, and the average of the detrimental bacteria was calculated as the same as (B). As shown in (B), if the average of beneficial bacteria was fixed to 40 ηg and the average of acne bacteria changed from 170 ηg to 210 η g, the average increase of detrimental bacteria was + 5.69, and if the average of beneficial bacteria was fixed at 60 ηg, the average increase of detrimental bacteria was + 2.76. In other words, the average increase of detrimental bacteria was about twice as low as beneficial bacteria were 1.5 times higher. In addition, if the average of acne bacteria was fixed at 170 ηg, and the average of beneficial bacteria changed from 40 ηg to 60 ηg, the average increase of detrimental bacteria was - 1.723. And if the average of acne bacteria was fixed at 210 ηg, the average increase of detrimental bacteria was - 4.653. In other words, the average increase of detrimental bacteria was about 2.7 times as low when the average increase of beneficial bacteria was 1.5 times higher, and when the average increase of acne bacteria was about 1.2 times higher.

However, as shown in (D), when the average of commensal bacteria was high as 450 ηg, it was different from the characteristics of (B) and (C). In particular, the average of detrimental bacteria was different when the average of acne bacteria was fixed and the average of beneficial bacteria was changed. If the average of beneficial bacteria was fixed at 40 ηg and the average of acne bacteria changed from 170 ηg to 210 ηg, the average increase of detrimental bacteria was + 6.12. When the average of beneficial bacteria was fixed at 60 ηg, the average increase of detrimental bacteria was + 3.19 and decreased by about 1.9 times. On the other hand, when the average of acne bacteria was fixed at 170 ηg, and the average of beneficial bacteria fluctuated from 40 ηg to 60 ηg, the average increase of detrimental bacteria was + 1.051. And when the average of acne bacteria was fixed at 210 ηg, the average increase of detrimental

bacteria was - 1.879. In other words, when the average of commensal bacteria was high and the average of acne bacteria was low, even if the average of beneficial bacteria was increased, the average increase of harmful bacteria was not decreased, but rather increased. In addition, when both the average of commensal bacteria and acne bacteria was high, increasing the average of beneficial bacteria resulted in a decrease in the average increase of detrimental bacteria.

These results complemented Figure 3, and the results of this study were verified empirically by using multiple regression models that beneficial bacteria, commensal bacteria, and acne bacteria that constituted skin microbiomes not only independently affected detrimental bacteria, but also had an effect on detrimental bacteria through interactions between beneficial bacteria and commensal bacteria, acne bacteria and commensal bacteria, and beneficial bacteria and acne bacteria.

4. Conclusion and Discussion

This study was conducted to empirically analyze the skin microbiomes across gender and age groups using bio big data, and to provide basic data analysis and information on the ecology of the skin microbiomes to the cosmetics manufacturing industry. The discussions of the research results are as follows.

First, the composition of the skin microbiomes was different across gender and age groups. The ratio of commensal bacteria was relatively high in both males and females, followed by acne bacteria in males, detrimental bacteria in females, and the distribution of detrimental bacteria was the lowest in males and females. In the age group of males, the 40s were different from their 20s and 30s, and females' age group was different from the distribution of the skin microbiomes in their 30s and 40s.

A recent study presented the composition of the skin microbiomes in 73 Korean women by dividing them into three groups: 10 ~ 29, 30 ~ 49, and 50 ~ 79, and analyzed that the distribution of all microbiomes in the forehead varied by age group, but the distribution of microbiomes in the hands was similar [32]. And it was suggested that the balance of the skin microbiomes should be considered according to age because the detrimental bacteria that contributed to maintaining skin health were affected by the increase of age. The case of Japan, which studied 37 women, reported that the distribution of microbiomes was diverse by dividing them into young people aged 21 ~ 37 and elderly people aged 60 ~ 76[33]. In addition, a study reported the characteristics of skin microbiomes in 73 Chinese women, young people aged 25 ~ 35, and elderly people aged 56 ~ 63, with different microbiomes distributed in the two groups, especially the structure of the skin microbiomes in the elderly was different from the younger, and the reason was the difference between the residential area and the age of the study[34]. Several pieces of research have limited studies on women as well as small samples, and the ranges of each age group were not the same, making it difficult to generalize the composition of skin microbiomes[32-34].

Whereas this study was designed to generalize the composition distribution of skin microbiomes compared to previous studies by analyzing 287 ordinary people living throughout Korea by gender and age group in consideration of regional differences in the skin microbiomes. Therefore, the differences in the composition and ratio of skin microbiomes by gender and age analyzed through this study are meaningful in that it provides basic data that can be used in the cosmetics industry in the future.

And as a result of analyzing the average difference in skin microbiomes by gender and age, there was no significant difference in commensal bacteria, acne bacteria, and beneficial bacteria, but only detrimental bacteria showed significant differences in average according to gender and age group. The average of detrimental bacteria was significantly higher in females than in males, and the age group showed a significant difference only in their 20s and 40s. As a related previous study, the effect of age and gender on the skin-associated microbial communities was studied in 71 people living in Shanghai, China, and the relative abundance of *Corynebacterium* among detrimental bacteria was significantly higher in males than females[35]. On the other hand, the relative abundance of *Staphylococcus* was about three times higher in females than males. Although the region and age groups of the study subjects were different compared to this study, it was

confirmed that there was a significant difference in the average of detrimental bacteria in males and females.

These results showed that the distribution of the skin microbiomes was different according to gender, and within gender, it was different according to age group. Therefore, it was empirically confirmed that the manufacture of cosmetics using skin microbiomes required a customized design according to gender and age. This could be confirmed again in the average analysis of the skin microbiomes by gender and age, especially since the difference between detrimental bacteria was significant, it was suggested that a design which can satisfy consumers' needs for healthy skin was essential to developing personalized cosmetics for detrimental bacteria.

Second, in the case of a multiple regression analysis that was controlled for gender and age with differences in skin microbiome distribution, beneficial bacteria had a negative effect on detrimental bacteria, while commensal bacteria and acne bacteria had a positive effect on detrimental bacteria.

In the correlation of the skin microbiomes, detrimental bacteria showed the relatively highest correlation coefficient of + 0.739, acne bacteria was + 0.518 and beneficial bacteria was + 0.121, showing a weak positive correlation. When analyzed by gender, the correlation between detrimental bacteria showed a relatively stronger correlation coefficient for females than males. Especially, the correlation between beneficial bacteria and detrimental bacteria was not significant in both male and female groups.

Since the results of this correlation analysis were based on the characteristics of only two variables, so the ecology of the skin microbiomes was empirically confirmed through the results of multiple regression analysis that controlled gender and age, which were influencing factors of the skin microbiomes.

In Table 6, Model 1 designed gender and age as control variables and analyzed the effects of beneficial bacteria, commensal bacteria, and acne bacteria on detrimental bacteria as independent variables. As a result, in the correlation analysis, beneficial bacteria and detrimental bacteria were not significant in both males and females, but in the multiple regression analysis, they were significant as a negative influence. In other words, the increase in the influence of beneficial bacteria in the same gender and age group showed a decrease in detrimental bacteria. So, in order to reduce detrimental bacteria that cause skin troubles and inflammation, it is necessary to design customized cosmetics to help form a skin environment that can increase the distribution of beneficial bacteria suitable for gender and age groups.

And another customized design that can reduce the negative effects of detrimental bacteria on the skin is to provide a skin environment in which acne bacteria are reduced. Since the increase of acne bacteria has a positive effect on detrimental bacteria, customized cosmetics designed for reducing acne bacteria according to gender and age group characteristics may also be considered.

There have been studies that reduce the influence of harmful bacteria and form a balance of healthy skin, suggesting the possible ways to reduce acne bacteria or aerobic detrimental bacteria that worsen skin inflammation [31,36]. And a study reported positive results in improving collapsed skin homeostasis using *Lactobacillus* ferments[10]. Another study introduced early studies on atopic dermatitis that inhibited pathogens by rebuilding the skin microbiomes or supplementing with beneficial bacteria[11]. However, these studies reported the results of clinical trials on small samples, so it was difficult to generalize and reported limitations that continuous research should be conducted in the future. It also proposed a method of controlling the microbiome, but most previous studies mentioned deficiencies that were still in the experimental stage or did not have sufficient evidence. In this regard, this study is meaningful in that it can help to understand the ecology of the skin microbiomes by empirically presenting the effects of beneficial bacteria, detrimental bacteria, and acne bacteria on harmful bacteria.

Third, in the skin microbiomes, beneficial bacteria, commensal bacteria, and acne bacteria not only independently affected detrimental bacteria, but also affected each other through interaction.

Model 2 of Table 6 designed gender and age as control variables affecting detrimental bacteria as shown in Model 1, and designed beneficial bacteria, commensal bacteria, and acne bacteria as independent variables. At the same time, the interaction terms of beneficial bacteria and commensal

bacteria, acne bacteria and commensal bacteria, and beneficial bacteria and acne bacteria were analyzed together. As a result, Model 2 had a 15.9%p higher explanatory power than Model 1, and all interaction terms showed statistically significant results. These results meant that beneficial bacteria, commensal bacteria, and acne bacteria in the skin microbiomes not only affected detrimental bacteria independently, but also constructed and affected skin ecology through interaction with each other.

The skin microbiomes form a balance, they cause problems such as weakening of the skin's immune system or causing inflammation[14,22]. Commensal bacteria show the characteristics of intermediate bacteria to help the growth of detrimental bacteria, but also increase the effect of detrimental bacteria [12]. In other words, skin microbiomes do not only exchange their effects, but also affect and balance through interactions with each other.

Figure 3 and Table 7 were the results of an empirical analysis of this, and there were interactions among the skin microbiomes where the average of detrimental bacteria was different when beneficial bacteria and acne bacteria changed according to the level of the commensal bacteria. When the level of the commensal bacteria was average, the average reduction rate of the relatively highest detrimental bacteria was shown through interactions between beneficial bacteria and acne bacteria. However, when commensal bacteria were higher than the average and acne bacteria were lower, detrimental bacteria were increased even if the beneficial bacteria were increased.

This result means that only the beneficial bacteria should not be increased simply for customers who are recognized as having problems with the skin when designing personalized cosmetics. In fact, women in their 20s and 40s living in Jeolla-do Province were asked to consult personalized cosmetics due to skin trouble. As a result of the PCR analysis of these people, the ecology of the skin microbiomes was different from the distribution of ordinary people. For them, customized cosmetics consisting of *Lactobacillus* which were beneficial bacteria and natural extracts, were applied to their skin, but it got worse. Therefore, personalized cosmetics should consider not only the independent effects of the skin microbiomes but also their interaction mechanisms.

Previous studies on the interaction of the skin microbiomes have been described only by theoretical content[12,22], and reported that *S. aureus* interacted with other microorganisms concerning certain diseases[7,21,24]. In addition, a couple of studies reported through clinical trials that there were many interactions between skin bacteria and *S. aureus*[36,37], and another study claims that interactions exist in other microorganisms besides *S. aureus*[38]. These previous studies have remained in theoretical content or have only revealed that certain microorganisms such as *S. aureus* interact with other microorganisms through small samples of clinical trials. Therefore, this study can contribute to preparing basic data on the personalized cosmetics industry by empirically analyzing and understanding the ecological characteristics and interactions of the skin microbiomes.

The implications of this study are as follows: to reduce detrimental bacteria that cause skin inflammation or trouble, this study is suggested that the skin environment should not be limited to increasing the influence of beneficial bacteria, but also the characteristics of the skin microbiomes that interact with each other between beneficial bacteria, commensal bacteria and acne bacteria. In addition, this study is meaningful in that present the results of empirically analyzing the characteristics of the skin microbiomes affecting detrimental bacteria on the skin using bio big data, as well as that derive the functional characteristics of interactions within the skin microbiomes that are still unexplored. Nevertheless, this study has limitations in that various internal and external factors affecting the skin microbiomes have not been considered. In terms of the personalized cosmetics industry, the types of commensal bacteria are very diverse and difficult to control, and related laws and application methods are very limited in utilizing beneficial bacteria. There should be supplemented more clinical and empirical studies on the interactions among the skin microbiomes in the future, and various case studies are needed in the personalized cosmetics industry.

References

1. J. G. Choi, S. S. Park S. J. Luy, C. Y. Luy, J. H. Lee, G. E. Woo, Y. J. Na, J. A. Lee and H. D. Lee, Statistics

on technology patents related to the 4th industrial revolution, The Convergence Technology Examination Bureau in Korean Intellectual Property Office (2022). Accessed: April. 25, 2022. [Online] Available: https://www.korea.kr/archive/expDocView.do?docId=39228
2. H. J. Do, COSMAX, Discovery of the 2nd generation skin microbiome, The Herald Economy (2022). Accessed: May. 5, 2022. [Online] Available: http://news.heraldcorp.com/view.php?ud=20220324000581
3. G. S. Kim and S. M. Kang, The skin safety and effects of the fermented broth of *Lactobacillus rhamnosus* for improving the neck skin, *J. Kor. Soc. Cosm.*, **23**(1), 101 (2017).
4. M. K. Song, Sangyoyeon introduces microbiome R&D trends, The Today Energy (2022). Accessed: April. 20, 2022. [Online] Available: http://www.todayenergy.kr/news/articleView.html?idxno=246864&lang=en
5. S. K. Jeong, Skin microbiome for cosmetic application, *The Journal of Skin Barrier Research*, **21**(1), 30 (2019).
6. Ministry of Food and Drug Safety. Research and analysis of global market trends in customized cosmetics (2022). Accessed: May. 11, 2022. [Online] Available: https://mfds.go.kr/files/upload/etc/e-Book/guideLine_6/index.html
7. M. H. Kim, D. I. Suh, S. Y. Lee, Y. K. Kim, Y. J. Cho, and S. H. Cho, Microbiome research in food allergy and atopic dermatitis, *Allergy Asthma Respir. Dis.*, **4**(6), 389 (2016).
8. K. H. Jeong, Skin and intestinal microbiome in psoriasis, *J. Kor. Soc. Psor.*, **16**(1), 8 (2019).
9. K. S. Cho, Research trends and prospects of human microbiome, *Gwang-Jang*, **221**, 255 (2021).
10. E. A. Grice and J. A. Segre, The skin microbiome, *Nat. Rev. Microbiol.*, **9**, 244 (2011).
11. B. Y. Kim, Microbiome research and industrial utilization, *BT News*, **28**(2), 16 (2018).
12. K. Skowron, J. Bauza-Kaszewska, Z. Kraszewska, N. Wiktorczyk-Kapischke, K. Grudlewska-Buda, J. Kwiecińska-Piróg, E. Wałecka-Zacharska, L. Radtke, and E. Gospodarek-Komkowska, Human skin microbiome: impact of intrinsic and extrinsic factors on skin microbiota, *Microorganisms*, **9**(3), 543 (2021).
13. S. Zhang and E. Duan, Fighting against skin aging: the way from bench to bedside, *Cell Transplantation*, **27**(5), 729 (2018).
14. P. B. Eckburg, E. M. Bik, C. N. Bernstein, E. Purdom, L. Dethlefsen, M. Sargent, S. R. Gill, K. E. Nelson, and D. A. Relman, Diversity of the human intestinal microbial flora, *Science*, **308**(5728), 1635 (2005).
15. E. A. Grice, H. H. Kong, G. Renaud, A. C. Young. NISC Comparative Sequencing Program, G. G. Bouffard, R. W. Blakesley, T. G. Wolfsberg, M. L. Turner, and J. A. Segre, A diversity profile of the human skin microbiome, *Genome Res*, **18**(7), 1043 (2008).
16. J. C. Clemente, E. C. Pehrsson, M. J. Blaser, K. Sandhu, Z. Gao, B. Wang, M. Magris, G. Hidalgo, M. Contreras, O. Noya-Alarcon, O. Lander, J. Mcdonald, M. Cox, J. Walter, P. L. Oh, J. F. Ruiz, S. Rodriguez, N. Shen, S. J. Song, J. Metcalf, R. Knight, G. Dantas, and M. G. Dominguez-Bello, The microbiome of uncontacted Amerindians, *Sci. Adv.*, **1**(3), e1500183 (2015).
17. S. M. Choi, S. H. Cho, and H. N. Yi, Human microbiome studies in Korea, *Allergy Asthma Respir. Dis.*, **4**(5), 311 (2016).
18. G. S. Kim and S. M. Kang, The effects of the fermented broth of *Lactobacillus rhamnosus* for improving the facial skin of middle-aged women as the skin barrier-recovering cosmetic material, *J. Kor. Soc. Cosm.*, **22**(5), 1004 (2016).
19. M. H. Leung, D. Wilkins, and P. K. Lee, Insights into the pan-microbiome: skin microbial communities of Chinese individuals differ from other racial groups, *Sci. Rep.*, **5**(11845), 1 (2021).
20. M. H. Kim, D. I. Suh, S. Y. Lee, Y. K. Kim, Y. J. Cho, and S. H. Cho, Microbiome research in food allergy and atopic dermatitis, *Allergy Asthma Respir. Dis.*, **4**(6), 389 (2016).
21. H. Kim, K. Myoung, H. G. Lee, E. J. Choi, T. Park, and S. An, *Lactobacillus plantarum* APsulloc 331261 fermented products as potential skin microbial modulation cosmetic ingredients, *J. Soc. Cosmet. Sci. Korea*, **46**(1), 23 (2020).
22. A. L. Byrd, Y. Belkaid, and J. A. Segre, The human skin microbiome, *Nat. Rev. Microbiol.*, **16**, 143 (2018).

23. P. Y. Ong, T. Ohtake, C. Brandt, I. Strickland, M. Boguniewicz, T. Ganz, R. L. Gallo, and D. Y. M. Leung, Endogenous antimicrobial peptides and skin infections in atopic dermatitis, *N. Engl. J. Med.*, **347**(15), 1151 (2002).
24. H. H. Kong, J. Oh, C. Deming, S. Conlan, E. A. Grice, M. A. Beatson, E. Nomicos, E. C. Polley, H. D. Komarow, NISC Comparative Sequence Program, P. R. Murray, M. L. Turner, and J. A. Segre, Temporal shifts in the skin microbiome associated with disease flares and treatment in children with atopic dermatitis, *Genome Res.*, **22**(5), 850 (2012).
25. P. L. J. M. Zeeuwen, M. Kleerebezem, H. M. Timmerman, and J. Schalkwijk, Microbiome and skin diseases, *Curent Opinion in Allergy and Clinical Immunology*, **13**(5), 514 (2013).
26. K. Findley, J. Oh, J. Yang, S. Conlan, C. Deming, J. A. Meyer, D. Schoenfeld, E. Nomicos, M. Park, NISC Comparative Sequencing Program, H. H. Kong, and J. A. Segre, Topographic diversity of fungal and bacterial communities in human skin, *Nature*, **498**(7454), 367 (2013).
27. P. U. Giacomoni, T. Mammone, and M. Teri, Gender-linked differences in human skin, *J. Dermatol. Sci.*, **55**(3), 144 (2009).
28. R. Juge, P. Rouaud-Tinguely, J. Breugnot, K. Servaes, C. Grimaldi, M. P. Roth, H. Coppin, and B. Closs, Shift in skin microbiota of Western European women across aging, *J. App. Microbiol.*, **125**(3), 907 (2018).
29. Y. J. Park and H. K. Lee, The role of skin and orogenital microbiota in protective immunity and chronic immune-mediated inflammatory disease, *Front. Immunol.*, **8**(1955), 1 (2018).
30. M. Kim, T. Park, J. I. Yun, H. W. Lim, N. R. Han, and S. T. Lee, Investigation of age-related changes in the skin microbiome of Korean women, *Microorganisms*, **8**(10), 1581 (2020).
31. M. R. Williams and R. L. Gallo, Evidence that human skin microbiome dysbiosis promotes atopic dermatitis, *J. Invest. Dermatol.*, **137**(12), 2460 (2017).
32. S. M. Han, K. G. Lee, J. H. Yeo, W. T. Kim, and K. K. Park, Antimicrobial property of honeybee (*Apis mellifera* L.) venom against *Propionibacterium acnes* and aerobic skin flora, *Kor. J. Pharmacogn.*, **40**(3), 173 (2009).
33. N. Shibagaki, W. Suda, C. Clavaud, P. Bastien, L. Takayasu, E. Iioka, R. Kurokawa, N. Yamashita, Y. Hattori, C. Shindo, L. Breton, and M. Hattori, Aging-related changes in the diversity of women's skin microbiomes associated with oral bacteria, *Sci. Rep.,* **7**(10567), 1 (2017).
34. H. J. Kim, J. J. Kim, N. R. Myeong, T. Kim, D. Kim, S. An, H. Kim, T. Park, S. I. Jang, H. Y. Jae, I. Kwack, and J. S. Woo, Segregation of age-related skin microbiome characteristics by functionality, *Sci. Rep.*, **9**(16748), 1 (2019).
35. S. Ying, D. Zeng, L. Chi, Y. Tan, C. Galzote, C. Cardona, S. Lax, J. Gilbert, and Z. Quan, The influence of age and gender on skin-associated microbial communities in urban and rural human populations, *PloS one*, **10**(10), e0141842 (2015).
36. J. Y. Yu, S. H. Park, I. A. Hwang, S. J. Jo, C. H. Heo, S. U. Yun, and G. C. Park, A clinical study on the effect of a cream containing *Ramulus mori* extract and tea tree oil on acne vulgaris and aerobic skin flora, *K. J. Dermatol.*, **41**(9), 1136 (2003).
37. C. von Eiff, K. Becker, K. Machka, H. Stammer, and G. Peters, Nasal carriage as a source of *Staphylococcus aureus* bacteremia, *N. Engl. J. Med.*, **344**(1), 11 (2001).
38. C. Weidenmaier, C. Goerke, and C. Wolz, *Staphylococcus aureus* determinants for nasal colonization, *Trend. Microbiol.*, **20**(5), 243 (2012).