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Lion Corporation

Metagen, Inc.

Hiyoshi Oral Health Clinics

Lion Discovers that Oral Microbiomes*¹ of Those with Dental Caries and Periodontal Disease Differ from Those with Healthy Oral Condition,*² Even After Dental Treatment

: Aiming to Propose Preventive Methods that Take into Account the Oral Microbiome

Lion Corporation (President: Masayuki Takemori, hereinafter “Lion”) announces that, in collaboration with Metagen, Inc. (President and CEO: Shinji Fukuda, hereinafter “Metagen”) and Hiyoshi Oral Health Clinics (Chairman: Takashi Kumagai, hereinafter “Hiyoshi Clinics”), we have investigated the oral microbiome of people suffering from dental caries and periodontal disease before and after dental treatment as part of our efforts to promote preventive dentistry habits.

Through the results of this study, we clarified that the relative abundance ratio of oral bacteria related with dental caries and periodontal disease was higher even after dental treatment than that of healthy oral conditions, including a lower relative abundance ratio of nitrate-reducing bacteria, which may help prevent dental caries and periodontal disease. The results of this study were published in the online scientific journal “mSystems”, circulated by the American Society for Microbiology on September 12, 2023.

*1 Refers to groups of bacteria that live in the oral cavity. It is thought that individual bacteria form unique groups influenced by the person’s constitution, physical condition, diet, and lifestyle.

*2 Refers to someone who had no newly diagnosed decayed teeth over the past 5 years and no probing pocket depth deeper than 4 mm.

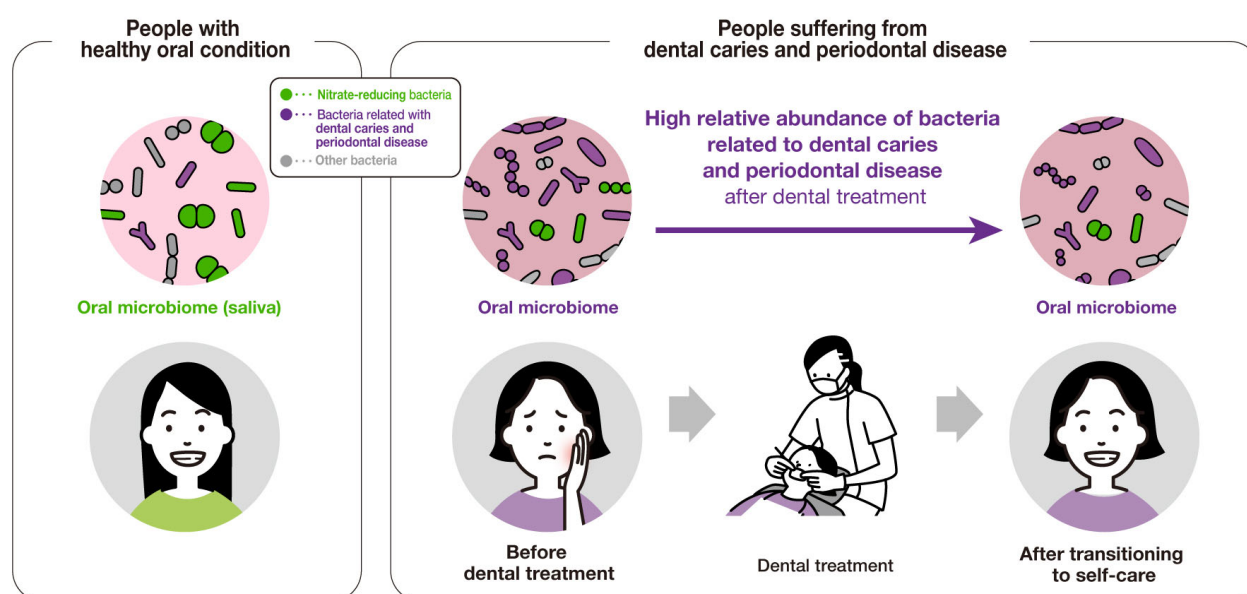


Figure1. Overview of Study

■ Research Background

Oral diseases, such as dental caries and periodontal disease, are caused by bacteria in the oral cavity, and can even lead to tooth loss. Recently, the oral microbiomes of those with these diseases has been found to be different from that of people with healthy oral conditions^{*3}, and it is thought that the oral microbiome characteristics of people with healthy oral conditions are important for prevention of these diseases. Also, these diseases have a high risk of recurrence.

Hence, we believe that understanding the oral microbiome after dental treatment-associated disease remission is necessary to prevent disease recurrence. Therefore, Lion collaborated with Metagen, which has its strengths in the analysis of microbiome, and Hiyoshi Clinics, which specializes in preventive dentistry, to investigate the oral microbiome before and after dental treatment.

^{*3} M Kilian *et al.* :The oral microbiome-an update for oral healthcare professionals, British Dental Journal, 221(10):657-666,2016.

■ Major Research Content

- Eligible subjects: People who met the relevant criteria among those who visited Hiyoshi Clinics from February 2018 to December 2020.
- Number of subjects: 12 people in the dental caries group,^{*4} 16 people in the periodontal disease group,^{*5} 22 people of the group with healthy oral conditions (hereinafter, the “healthy group”).^{*6}
- Main measurement items: Microbiome in saliva.
- Saliva collection: 3 times total, before dental treatment, after completion of dental treatment,^{*7} and after transitioning to self-care.^{*8}
- Dental treatment: Conducted in line with Hiyoshi Clinics’ treatment policies.^{*9}

^{*4} People had teeth requiring treatment.

^{*5} People had bleeding from probing pocket depth deeper than 4 mm.

^{*6} Refers to someone who had no newly diagnosed dental caries over the past 5 years and no probing pocket depth deeper than 4 mm.

^{*7} When the dentist determines that treatment is complete based on the patient’s oral condition and saliva test results.

^{*8} 3.5 months on average after the dentist determines that treatment is complete.

^{*9} Comprehensive treatment policies that include making initial risk assessments, providing guidance on lifestyle habits such as diet, and creating maintenance programs customized for each patient. (<https://www.hiyoshi-oral-health-center.org/naiyou/mtm/>)

■ Major Research Results

(1) Clarified that the oral microbiome of the dental caries and periodontal disease groups is different from that of the healthy group even after dental treatment

By comparing the oral microbiomes of the dental caries and periodontal disease groups and the healthy group, we found that the oral microbiome before dental treatment, after dental treatment, and after transiting to self-care of those in the dental caries and periodontal disease groups was significantly different from that of those in the healthy group.

(2) Dental caries and periodontal disease*¹⁰ groups had a higher abundance of bacteria related with dental caries and periodontal disease than the healthy group, even after dental treatment

We found that there were bacterial species that were in significantly higher abundance in the oral microbiomes of each disease group when compared to the healthy group. The relative abundance ratio of bacteria related to dental caries was higher in the dental caries group than in the healthy group, both before dental treatment and after transitioning to self-care. In the periodontal disease group, the relative abundance of bacteria related to periodontal disease was higher than in the healthy group before dental treatment to after transitioning to self-care (Figure 2).

*¹⁰ Lamont RJ *et.al.*: The oral microbiota: dynamic communities and host interactions, Nat Rev Microbiol,16(12)745-759, 2018.

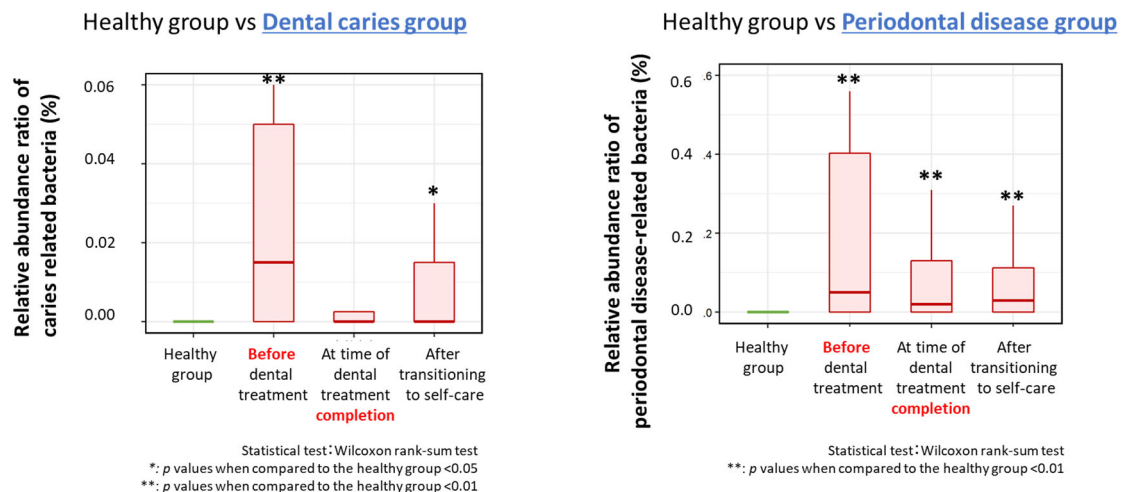


Figure 2a. Comparison of bacteria related to dental caries between the healthy and dental caries groups

(Comparison of the relative abundance ratio of *Bifidobacterium dentium*, a prominent species of caries-related bacteria)

Figure 2b. Comparison of periodontal disease-related bacteria between the healthy and periodontal disease groups

(Comparison of the relative abundance ratio of *Porphyromonas gingivalis*, a prominent species of periodontal disease-related bacteria)

(3) The relative abundance ratio of nitrate-reducing bacteria thought to contribute to the prevention dental caries and periodontal disease remains low even after dental treatment

We found that there were bacterial species that had significantly lower abundance ratio in the oral microbiome of each disease groups when compared to the healthy group. The relative abundance ratio of nitrate-reducing bacteria in both oral disease groups tended to be lower than in the healthy group both before dental treatment, and also after dental treatment was complete and the transitioning to self-care. It has been reported that metabolites produced by nitrate-reducing bacteria have the potential to inhibit acidification in the oral cavity and suppress the growth of bacteria related to dental caries and periodontal disease.*¹¹ The results suggest that increasing the abundance ratio of nitrate-reducing bacteria in the oral cavity may help prevent dental caries and periodontal disease.

*¹¹ Rosier BT *et.al.*: The Importance of Nitrate Reduction for Oral Health, J Dent Res, 101(8):887-897, 2022.

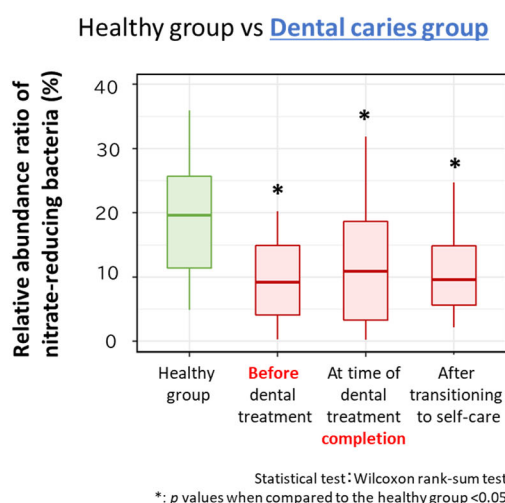


Figure 3a. Comparison of nitrate-reducing bacteria between the healthy and dental caries groups

(Comparison of the relative abundance ratio of *Neisseria flavescens*, a prominent species of nitrate-reducing bacteria)

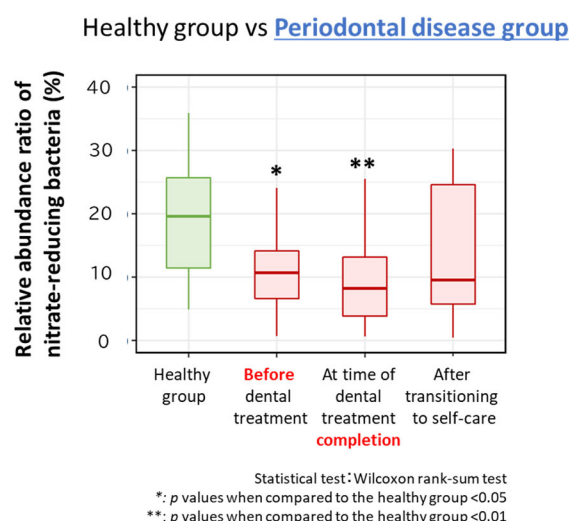


Figure 3b. Comparison of nitrate-reducing bacteria between the healthy and periodontal disease groups

(Comparison of the relative abundance ratio of *Neisseria flavescens*, a prominent species of nitrate-reducing bacteria)

Based on the above results, we found that the oral bacterial microbiomes of the dental caries and the periodontal disease groups are different from that of the healthy group even after dental treatment, which suggests that these conditions may lead to the easy recurrence of dental caries and periodontal disease. We believe that it will be important to devise new preventive methods that take the oral microbiome into account in order to prevent dental caries and periodontal disease, and we are proceeding with research activities.

■ Future Outlook

Lion is steadily implementing all of its corporate activities based on its basic approach to the oral health field set out in the Lion Oral Health Initiative,^{*12} and this research was conducted as part of the initiative. Going forward, we will continue to focus on the oral microbiome and accelerate research on the relationships between the microbiome and oral conditions at various life stages, with the aim of helping to improve consumer health, starting from the mouth.

^{*12} The collective name for activities in the oral health field aimed at realizing the Company's long-term strategic framework, Vision2030. For an overview, please see the announcement published on August 8, 2022.
(https://doc.lion.co.jp/uploads/tmg_block_page_image/file/8265/20220808a.pdf)

Metagen will continue working to elucidate the relationships between the human microbiome, health, and disease prevention, and strive to eradicate disease through frontier science.

Hiyoshi Clinics will continue developing new dental care methods to enhance the health of those in its communities and further improve the quality of life of many citizens. To this end, they will further expand their medical care services, improve the comprehensive capabilities of their clinics to provide more specialized dental care, continue to collect clinical data every day to pursue new knowledge to contribute to the future health of the community.

【Published Paper】

- Title : Dysbiosis of Oral Microbiome Persists After Dental Treatment-Induced Remission of Periodontal Disease and Dental Caries
- Authors : Yama K, Nishimoto Y, Kumagai K, Jo R, Harada M, Maruyama Y, Aita Y, Fujii N, Inokuchi T, Kawamata R, Sako M, Ichiba Y, Tsutsumi K, Kimura M, Murakami S, Kakizawa Y, Kumagai T, Yamada T, Fukuda S
- Publication: mSystems
- DOI : 10.1128/msystems.00683-23

【Company Overview】

◆ Lion Corporation

The Lion Group's management vision, "Becoming an advanced daily healthcare company," is driven by its purpose, "Make a difference in everyday lives by redesigning habits: ReDesign," and the Group is carrying out corporate activities accordingly. Going forward, we will continue providing new value unique to the Company as an oral care manufacturer.

- Corporate website : <https://www.lion.co.jp/en/>
- R&D website : <https://www.lion.co.jp/en/company/rd/>
- Related R&D case studies : <https://www.lion.co.jp/en/rd/basic/analysis/case01.php>

◆ Metagen, Inc.

Metagen, Inc.'s mission is to make gut environment-based healthcare a new standard, and our group is striving to achieve zero disease through its "Gut Design" approach. Under the slogan "Beyond Science, Beyond Technology, With Society", we will comprehensively promote the gut environment research and its social implementation to satisfy the desires of each and every individual.

◆ Hiyoshi Oral Health Clinics

The medical philosophy of Hiyoshi Oral Health Clinics is to make the oral health of the Sakata City community the best in the world, and its ultimate goal is KEEP28 (maintaining 28 healthy permanent teeth for a lifetime). Through the dental care the organization provides, Hiyoshi Clinics will continue its efforts in clinics, research, and education to support the oral health of both the residents of Sakata City and also that of people around the world.

- Hiyoshi Oral Health Clinics (Sakata) : <https://www.hiyoshi-oral-health-center.org>
- Hiyoshi Oral Health Clinics Shiodome : <https://www.hiyoshi-shiodome.com>