December 21, 2017

FINAL REPORT

Type of Award: Orthodontic Faculty Development Fellowship Award (OFDFA) - A126712

Principal Investigator: Andrew Jheon

Title of Project: Alterations in the subgingival microbiome during orthodontic treatment

Period of AAOF Support: 07-01-15 to 06-30-16 (1st NCE to 6-30-17; 2nd NCE to 12-31-17)

Amount of Funding: \$20 000

Summary/Abstract of Completed Project Results (250 word maximum):

Introduction: To evaluate changes in the subgingival microbiome before and during fixed appliance orthodontic treatment using 16S rRNA-based high-throughput sequencing. Methods: Sixteen patients (10 females and 6 males; ages 12y1m-33y3m) were included in this study. Subgingival microbial samples were collected from 4 teeth of each subject at three different times before and during full-fixed appliance treatment; two patients received clear aligner therapy (i.e., Invisalign). DNA was extracted from the samples, and 16S rRNA-based, next generation sequencing (NGS) was performed catalog global profiles of subgingival microbiome during orthodontic treatment. Results: The frequency of T forsythia, C rectus, and P nigrescens significantly increased after placement of orthodontic appliances. For the other species, the frequency tended to increase but no statistically significant differences were noted. The frequency of the change, representing microorganisms not existing at t0 but newly developing at t1 and t2, was higher at the molars than at the incisors. Although sample size was limited, there also appeared to be significant changes in patients receiving clear aligner therapy. Conclusion: Orthodontic treatment, whether utilizing full fixed appliances or clear aligners significantly alters subgingival microbial composition and consensus bacterial species that were altered were identified. Our experiments lay a solid foundation for further analysis of subgingival microbiome during orthodontic therapy.

Response to the following questions:

1. Were the original, specific aims of the proposal realized?

The original aim was "to profile changes in microbial composition during fixed orthodontic treatment using 16S rRNA next generation sequencing (NGS). Yes, this original aim has been realized. Our study was the first time that the oral microbiome was globally profiled during orthodontic treatment using NGS.

2. Were the results published?

No not yet. However, the manuscript is in preparation for submission to AJODO (I have attached the current draft). In addition, we will use our data as preliminary results to apply for NIDCR R21 funding for an expanded study.

a. If so, cite reference/s for publication/s including titles, dates, author or co-authors, journal, issue and page numbers.

Isaac Chen, Jennifer Chung, Yanjiao Zhao, Andrew H. Jheon (in preparation) Alterations in the subgingival microbiome during orthodontic treatment. AJODO.

- b. Was AAOF support acknowledged? Yes.
- c. If not, are there plans to publish? Yes, please see above.
- 3. Have the results of this proposal been presented? No
- a. If so, list titles, author or co-authors of these presentation/s, year and locations: N/A
- b. Was AAOF support acknowledged? N/A
- c. If not, are there plans to do so? If not, why not? No because we did not budget for any travel to conferences.
- 4. To what extent have you used, or how do you intend to use, AAOF funding to further your career?

I have applied for BRA funding for 2018 and hope to continue to apply for AAOF funding in the future. It is a valuable source of funding outside of the NIH for my research and laboratory.

Accounting for Project; i.e., any leftover funds, etc.

There are no leftover funds. I apologize that this project took much longer than anticipated. This was largely due to this being my first foray into human microbiome research. Thus, I did not properly account for timing of human research approval and data analysis. Thank you for your patience.

Please return to AAOF via email attachment to aaofevp@aaortho.org

Alterations in the subgingival microbiome during orthodontic treatment

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Highlights

- 1) Our study is the first to globally profile bacteria using 16S rRNA-based, next generation sequencing (NGS) to catalog changes in subgingival microbiota during orthodontic treatment.
- 2) There are dramatic alterations in subgingival microbiome during full fixed appliance orthodontic treatment.
- 3) Although there is little consensus in microbial profile in teeth between subjects and within the same subject, there are several consensus bacterial species that are significantly changed during orthodontic treatment.
- 4) Although subjects were limited, the use of clear aligners (e.g., Invisalign) also showed significant changes in subgingival microbiome during treatment, perhaps surprising since a company-touted advantage of clear aligners is their perceived neutral effect on oral hygiene.
- 5) Our study builds a strong foundation to further analyze the role of subgingival microbiome in orthodontic tooth movement utilizing full fixed appliance and clear aligner therapies.

ABSTRACT

Introduction: To evaluate changes in the subgingival microbiome before and during fixed appliance orthodontic treatment using 16S rRNA-based high-throughput sequencing. **Methods**: Sixteen patients (10 females and 6 males; ages 12y1m-33y3m) were included in this study. Subgingival microbial samples were collected from 4 teeth of each subject at three different times before and during full-fixed appliance treatment; two patients received clear aligner therapy (i.e., Invisalign). DNA was extracted from the samples, and 16S rRNA-based, next generation sequencing (NGS) was performed catalog global profiles of subgingival microbiome during orthodontic treatment. Results: The frequency of T forsythia, C rectus, and P nigrescens significantly increased after placement of orthodontic appliances. For the other species, the frequency tended to increase but no statistically significant differences were noted. The frequency of the change, representing microorganisms not existing at t0 but newly developing at t1 and t2, was higher at the molars than at the incisors. Although sample size was limited, there also appeared to be significant changes in patients receiving clear aligner therapy. Conclusion: Orthodontic treatment, whether utilizing full fixed appliances or clear aligners significantly alters subgingival microbial composition and consensus bacterial species that were altered were identified. Our experiments lay a solid foundation for further analysis of subgingival microbiome during orthodontic therapy.

Introduction

In 2007, the National Institutes of Health (NIH) funded the human microbiome project (HMP) to map microbial makeup of healthy humans in an effort to better understand correlation of the microbiome with human health [1]. It was discovered that nearly everyone routinely carries pathogens and microorganisms known to cause illnesses. However, in healthy individuals pathogens cause no disease and simply coexist with their host and the rest of the microbiome [1]. Recently, Sender et al. [2] published a revised estimate of bacteria to human cell ratio to be nearly 1:1, an estimated 38 trillion bacteria to 30 trillion human cells. Every human body contains personalized microbiomes that are essential to maintain health, but also capable of eliciting disease. For example, there is a concentration of 10¹¹ bacteria/ml in the gut that provide both beneficial and harmful effects [2]. This gut flora enhances our immune system, helps absorb vitamins, and utilizes the food consumed. Conversely, the gut flora is also associated with obesity, intestinal and systemic inflammation, and even cancer and autism [3].

Bacterial diversity in dental plaque and oral biofilm is estimated to include at least 800 different species, consisting of a wide variety of gram-positive and gram-negative bacteria [4]. The number of species is expected to rise into the thousands with the application of next-generation sequencing (NGS) techniques [5, 6]. Within the oral cavity, there are three main types of surfaces for bacteria to colonize: the hard surfaces of teeth (i.e., supra- and sub-gingival), the soft tissues of the oral mucosa, and saliva [7]. Furthermore, the oral microbiome is extremely dynamic because of the oral cavity's

continual exposure with the external environment [8]. Saliva has a bacterial concentration of 10⁹ bacteria/ml whereas dental plaque has an equivalent bacterial concentration as the colon (i.e., 10¹¹ bacteria/ml) [2]. Studies have shown the oral cavity's microbiome to be a key source in the etiology of many oral and systemic diseases [9, 10].

Dental plaque is a dynamic, complex biofilm and a microbial ecosystem [11]. Bacterial diversity in dental plaque or oral biofilm is estimated to include at least 800 different species, consisting of a wide variety of gram-positive and gram-negative bacteria [4, 12-15]. The number of species is expected to rise into the thousands with the application of next-generation sequencing (NGS) [5, 6]. In addition, many species in kingdom Monera, also known as archaebacteria, have been associated with oral disease such as chronic periodontitis [16, 17]. On top of the staggering number of bacteria and archaebacteria, dental plaque comprises an assortment of micro-niches, metabolic functions, and a web of inter- and intra-species interactions that accumulates through sequential and ordered colonization by different bacterial strains and species [18]. Biofilms develop under a wide range of different conditions, environments, and factors (e.g., pH, availability of oxygen and nutrients, and interbacterial co-adhesion), and these differences all have been demonstrated to lead to compositional changes in oral biofilm [18]. Oral biofilm profiling has been hampered because it is extremely difficult to define normal microbiota because of individual variation [19]. However, dental health has been associated with the absence [20, 21] or elevation [22-24] of certain species.

The composition of subgingival microbiota can be influenced by several factors including oral hygiene regimens, dental restoratives, and orthodontic appliances. The etiology of gingivitis and periodontitis is microbial infection, resulting in an imbalance between the host and subgingival microorganisms [17]. Fixed appliances can change the subgingival microbial environment by increasing plaque accumulation and deepening the gingival sulcus [25, 26]. Orthodontic appliances generally increased the level of periodontopathogens in subgingival plaques [27-29], however, several studies have reported no significant difference [30] or a decrease in the level of periodontopathogens decreased during orthodontic treatment due to metal corrosion, which imposed toxic effects on the microorganism [31]. Thus, previous studies regarding changes in periodontopathogens during orthodontic treatment have been inconsistent.

All previous studies involving orthodontic full fixed appliances have used microbial analysis methods such as candidate-based PCR or bacterial culture methods [30], but these techniques suffer from selection biases or difficulty in growing cultures. 16S rRNA combined with next generation sequencing (NGS) has been used previously to profile the microbiome in health and disease including periodontal diseases [32-36], but never during orthodontic treatment. 16S rRNA/NGS catalogs all human and microbial DNA in each of the samples, which is then sorted to identify specific genetic signals found only in bacteria — such as the variable genes of bacterial ribosomal RNA called 16S rRNA that can identify the presence of different microbial species [1]. The purpose of this study is, for the first time, to track global changes in subgingival bacterial

composition before and after the placement of patients undergoing traditional full fixed appliance orthodontic treatment.

Materials and Methods

Subjects

Sixteen subjects (10 Females and 6 Males; ages 16.2-17.5) were selected among patients who arrived for orthodontic treatment at the Department of Orthodontics, University of California San Francisco. Six patients failed to complete the entire study. The study design was approved by the Ethics Committee (IRB 15-17868). Subjects were enrolled according to the following criteria: (1) no known systemic disease; (2) no use of antimicrobial, antifungal or anti-inflammatory drugs within 3 months before the baseline examination; (3) no craniofacial anomalies; (4) not-pregnant; and (5) would be undergoing treatment with fixed orthodontic appliances in the mandibular arch. Five subjects (4 female and 1 male; Age ??) not undergoing orthodontic treatment were selected as a control group.

Orthodontic bonding

All subjects received oral hygiene instructions by the same clinician in the form of both verbal instructions and an instructional video by Dolphin Aquarium (Patterson Technology). Subjects were instructed to continue their routine oral hygiene regiment. All subjects received orthodontic therapy with fixed buccal appliances. For orthodontic treatment, metal brackets (3M Unitek, GAC, Opal or GAC innovation) were bonded directly with composite resin (find brand of composite) onto incisors and premolars. Bands (Unitek and GAC) were cemented with polyacid-modified composite resin (Ultra Band-Lok) onto molars. The arch wires were tied using o-chains (find brand of o-chains) and in one case self-ligated.

Clear aligner therapy

Two patients received clear aligner trays (Invisalign) and treatment was administered following instruction from Invisalign.

Collection of subgingival microbiota

Subgingival crevicular fluid samples were collected from the straight buccal gingival crevice of the lower right first molar (30B), lower right central incisor (25B), lower left first premolar (21B) and the straight lingual of lower left first premolar (21L). Samples were collected at three different time points: before appliance placement (t0), 6 weeks (t1) and 12 weeks (t2) into treatment. The sampling sites were isolated using full mouth isolators (Dentsply, GAC) and the tongue held back with a mouth mirror when sampling tooth 21L. Two sterile paper points size 30 (Dentsply, GAC) were inserted into the gingival crevice and were left *in situ* for 60 s. These paper points were transferred immediately into Eppendorf tubes containing 250 mL of 0.9% saline solution (Baxter, Grainger) and frozen at -80 degrees F.

Periodontal Assessment

Immediately after sampling the sites, the probing depths (PD), GM-CEJ measurement, clinical attachment loss (CAL), and bleeding on probing (BOP) were measured and recorded in Axium periodontal charting system for all three time points. A sterilized periodontal probe (find brand) was used to measurement PD and GM-CEJ. BOP was observed and classified as present or not present after the periodontal measurements. All sampling and measurements were performed by the same clinician. Once all the

samples were collected, they were sent to Jackson Laboratory in Bar Harbor, ME for 16S rRNA gene sequencing and analysis.

16S rRNA gene sequencing: Metagenomic DNA from the sampled paper points was extracted using a PowerSoil ® DNA Isolation Kit. 16S rRNA V1-3 regions from the metagenomic DNA were amplified using primers 27F and 534R (27F:5'-AGAGTTTGATCCTGGCTCAG-3' and ATTACCGCGGCTGCTGG-3'). 534R: 5'-Sequencing reads were processed by removing the sequences with low quality (average qual <35) and ambiguous nucleotides (N's); chimeric amplicons were removed using UChime software. OTU was generated from the processed reads using an automated pipeline. Each OTU was classified from phylum to genus level using the most updated RDP classifier and training set. A taxonomic abundance table was generated with each row as bacterial taxonomic classification, each column as sample ID, and each field with taxonomic abundance. The abundance of a given taxon in a sample was presented as relative abundance (the read counts from a given taxon divided by total reads in the sample).

Statistical Analysis

Each sample was subsampled to the lowest number of read counts among samples in the dataset and rarefied to 5000 read counts. The abundance of a taxon in a sample was indicated as the relative abundance, which was calculated by dividing the number of reads for a taxon by the total read counts of the sample. Alpha diversity indices including richness and Shannon diversity were calculated as previously described. To

examine difference between control and bracket samples, we performed non-metric multidimensial scaling (NMDS) plots with Bray-curtis distance. Microbiome stability in both groups was determined by calculating the Bray-curtis dissimilarity from the same samples across different time points.

Results (in preparation)

We obtained a mean of 23,755 sequences per sample and aggregated to a total of 402 operational taxonomic units (OTUs). In our braces cohort, we observed that there was a significant increase in microbial richness between samples obtained before braces intervention and six weeks after intervention among three of the four sites. Richness stayed at a similarly high microbial richness by 12 weeks. This increase in richness was accompanied by the decrease in Streptococcus, the most abundant genera found across all samples. Similar trends were observed when looking at diversity among samples by Bray Curtis. Samples taken before bracket treatment clustered more tightly and differently compared to the two time points after, which clustered similarly among each other. In addition, over 30% of our bracket cohort developed periodontitis compared to 15% of our controls by six weeks of intervention. Overall samples that were associated with periodontitis had a lower relative abundance of Streptococcus. We also observed that there were fewer OTUs shared among the four sites at t0 relative to t1 and t2, suggesting that the whole oral microbiome becomes more uniformed with bracket treatment. Interestingly, the unique OTUs to each site over time did not remain the same, suggesting the uniqueness of each tooth's bacterial profile may be transient.

Discussion (in preparation)

One of the major problems associated with phase II, fixed appliance treatment is its long duration, on average of 29 months [37]. There is empirical, clinical evidence that increased bacterial plaque may decrease the rate of orthodontic tooth movement and overall treatment times. In addition, the microbiome has recently been shown to possess critical, much larger roles in health and disease, most notably with studies on the effects of the gut microbiome on obesity [38], intestinal cancer [39], diabetes [40], and autism [3]. Finally, the biological and mechanical changes induced by orthodontic treatment would likely lead to changes in subgingival dental plaque. Thus, the contribution of the microbiome, and specifically subgingival plaque, during orthodontic therapy is an exciting possibility that has yet to be tested. Ultimately, manipulation of the microbiome may be a therapeutic option to accelerate orthodontic tooth movement and shorten treatment times.

We observed significant microbial changes in the oral microbiome of patients with bracket intervention and the composition of these communities may contribute to periodontitis.

Acknowledgment

We would like to thank the American Association of Orthodontics Foundation for supporting this work through the Orthodontic Faculty Development Fellowship Award (OFDFA) to A.H.J..

Figure Legends

Figure 1. Subgingival microbial profiles pooled for control (no treatment), full fixed appliance (bracket), and clear aligner (invisalign) therapy. For all groups, OTU 1.streptococcus is the most common bacterial genus, followed by OTU 3. Streptococcus and OTU 2. Veillonella. In the bracket group, tooth 30B showed an overall decrease in OTU 1. Streptococcus, whereas 25B and 21B showed decreases at t1 followed by increases at t2 although not to the same levels at t0. 21L profile remained relatively the same at all three time points. In the control group, the levels of OTU 1. Streptococcus had similar abundance at all three time points for 30B and 25B, an overall decrease in 21B but an increase in 21L. The bracket group showed an overall decrease in OTU 3. Streptococcus from t0 to t2, whereas the control group showed an overall increase in OTU 3. Streptococcus, except for tooth 30B. OTU 2. Veillonella increased for bracket tooth 25B and 21L but decreased in 30B and 21B. In the control group, OTU 2. Veillonella increased in tooth 25B, decreased in 30B and 21B, and was barely present in 21L. The amount of "other" bacteria was higher on the bracket group than in the control group.

Figure. 2. Bray-Curtis, Richness, and Shannan diversity analyses. (A) The Bray-Curtis analysis provides a measure of community composition differences between samples based on OTU counts regardless of taxonomic assignment. Ordinations based on this metric demonstrated a clear separation between control, bracket, and Invisalign groups for tooth 30B. 25B and 21L were slightly more similar and 21B showed

significant separation between t0-t1 but were more similar at t1-t2. (B) The bracket group demonstrated a significant increase in microbiome richness from t0 to t2, whereas the control group only showed slight increases in microbiome richness from t0 to t2. (C) The Shannon diversity index (H) is commonly used to characterize species diversity in a community. At t0 both bracket and control groups started with similar diversity indexes. The bracket group showed an increase in diversity for tooth 30B and 21L at t1 and t2, but tooth 25B and 21B showed increases at t1 followed by decreases at t3. In the control group, tooth 30B and 21B showed minimal changes in diversity, 25B showed a decrease in diversity at t1 and an increase at t2, whereas tooth 21L showed a decrease in diversity at t2.

Figure 3. Non-metric Multidimensial Scaling (NMDS) ordination analysis. To evaluate the relationship between bacterial community composition and various environmental factors, we performed NMDS analysis to represent pairwise Bray-Curtis similarities between bacterial communities.

Figure 4. Global subgingival microbial profiles of individual subjects and teeth.

Figure 5. Venn diagram showing shared OTU or consensus bacterial species in all teeth and time.

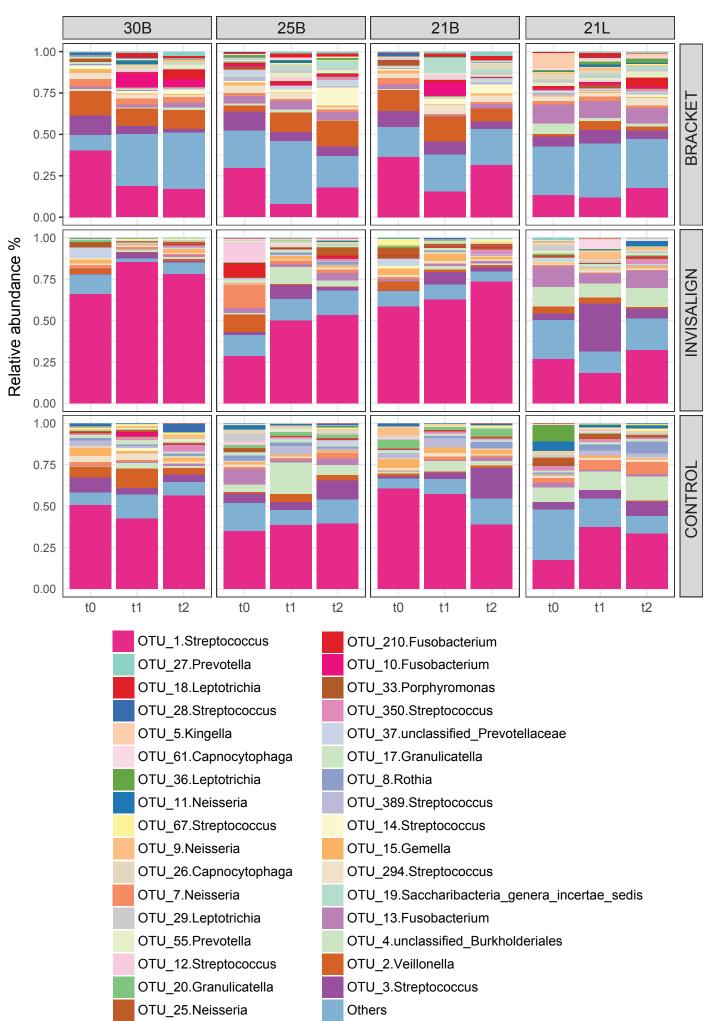
Figure 6. Venn diagram showing shared OTU or consensus bacterial species in all teeth at the three time points.

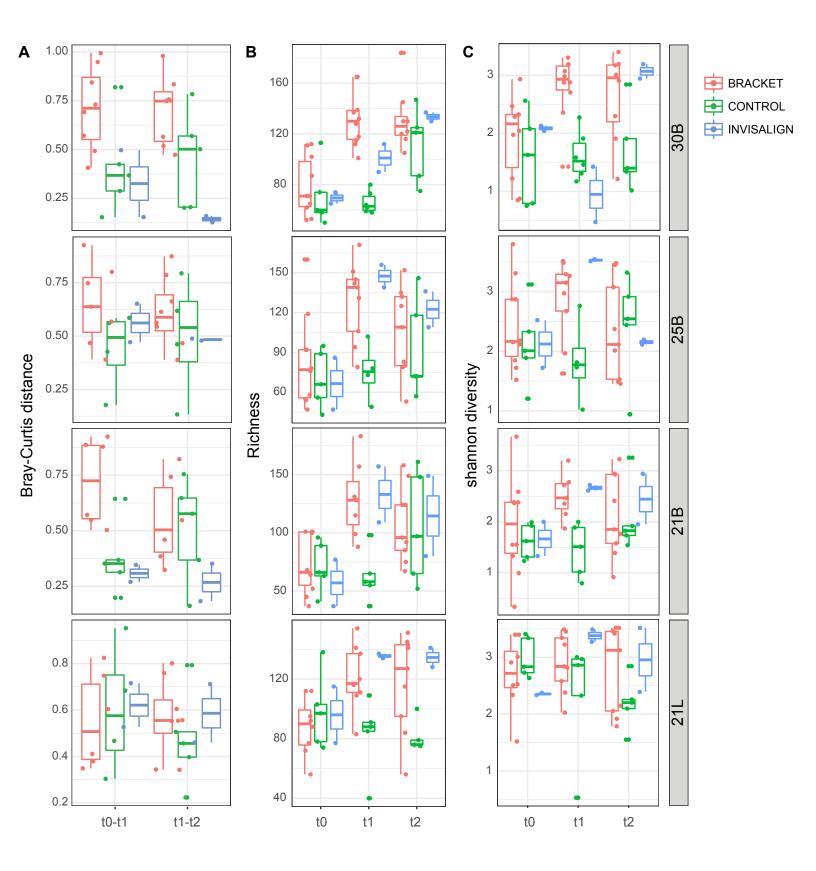
Supplemental Figure 1. Top 10 up- and down-regulated bacterial species for each tooth and subject at t0-t1 and t1-t2. Subjects are listed as A-O.

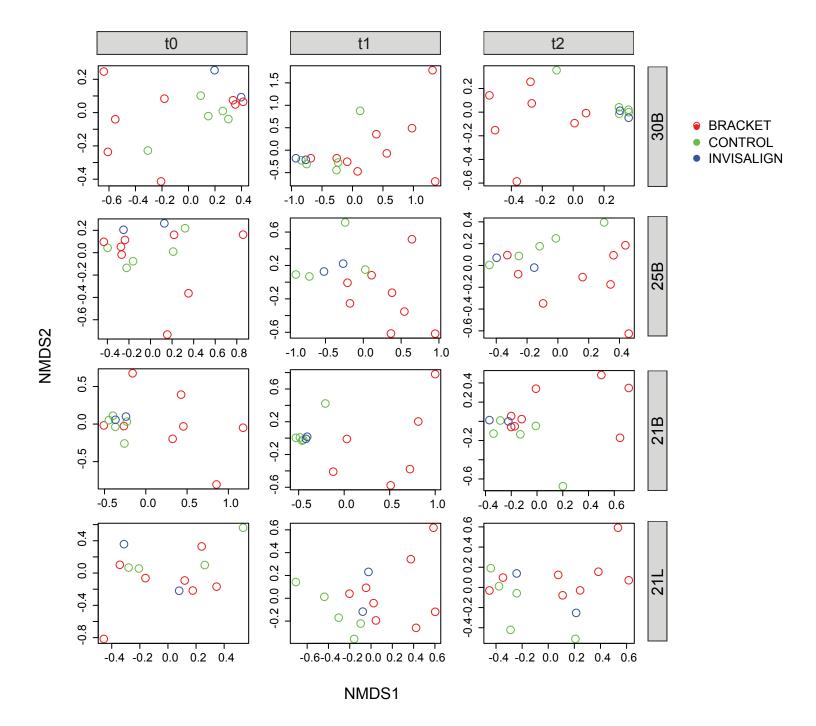
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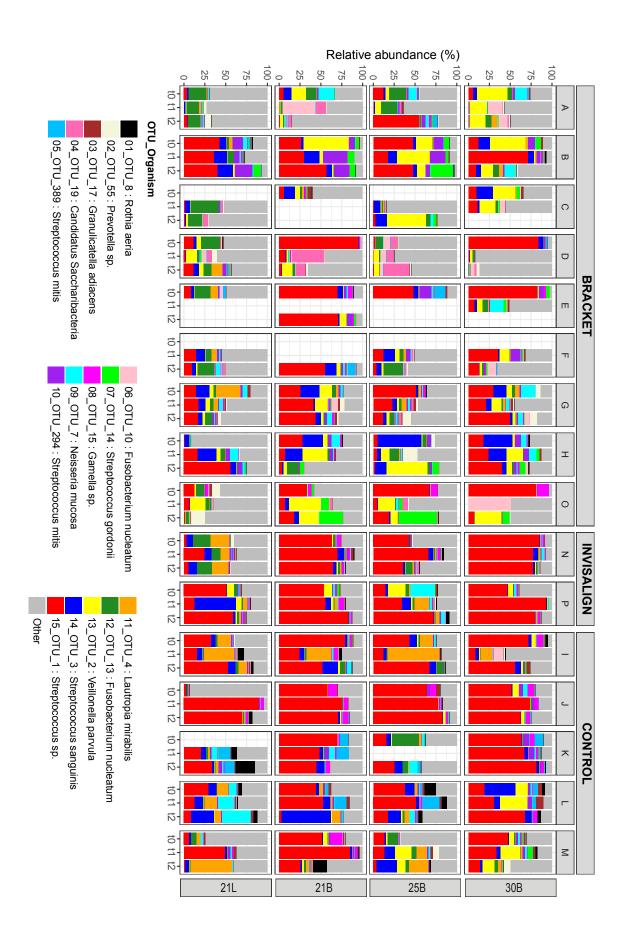
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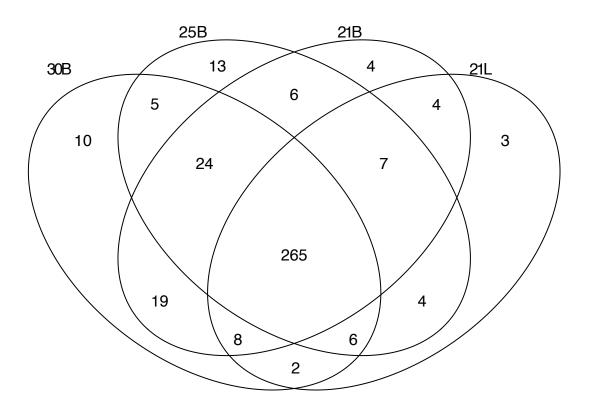




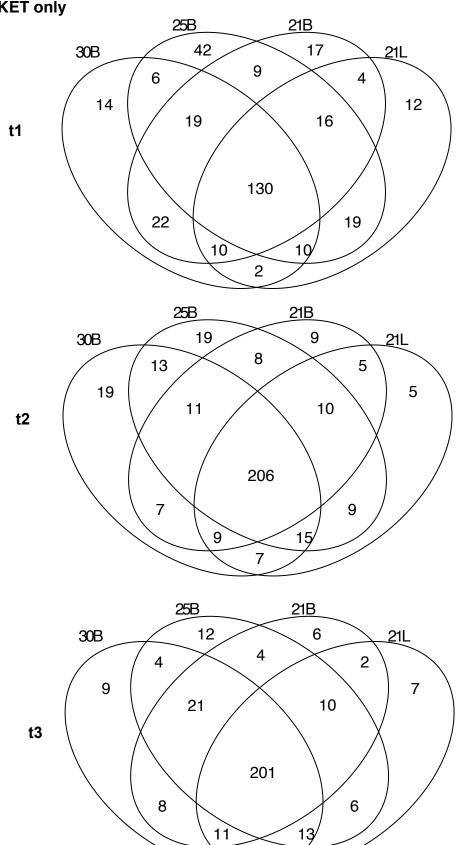




BRACKET only: Shared otu in among all teeth among all timepoints







6

SUPPLEMENTAL TABLE 1

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n 1: TP2-TP1 (308: pecies_nam diff_vi /eillonella di 16.82	259831 Veillonella di 40.853791 Streptococcu 17.7058	species_nam.diff_value species_nam.diff_va 44 Streptococcu 82.9024549 Streptococcu 74.994	llue 45688	species_nam.diff_value Neisseria mu 13.1436	species_nam.diff_v 02 Streptococcu 12.35	alue species_nam 40908 Streptococcu			nam diff_value specie coccu 7.82098631 Strept haer 2.31370845 Rothia		ptococcu 13.61068	87 Streptococcu	8.61394743 Streptoc	_nam.diff_value species coccu 81.0341082 Allopre a haer 13.2739518 Veillone	
leisseria mu 14.81 Porphyromoi 12.04 Pusobacteriu 4.714	164241 Streptococu 6.44145123 Veilonella di 7.125787 464914 Streptococu 5.76797741 Streptococu 4.149467 431675 Streptococu 5.26830328 Streptococu 3.019769	53 Streptococcu 4.00825549 Streptococcu 4.714; 74 Streptococcu 1.90093417 Gemeila hae; 2.585; 772 Streptococcu 1.53160982 Streptococcu 1.0536 05 Gemeila haei 0.69519878 Streptococcu 0.3367	66066	Streptococcu 9.146208 Streptococcu 8.494460 Prevotella lot 4.11688	13 Rothia mucil: 2.596 103 Haemophilus 2.118	18379 Streptococcu	7.41907452 Neisseria mu 3.66065609 Streptococcu 1.95524658 Gemella haei	4.36671736 Capnoc 3.56289376 Cardiob	ytoph 0.80382359 Strept acter 0.61916142 Actino	rcoccu 1.69454703 Ger nyces 1.41212253 Por	phyromo: 1.705409	44 Gemella haei 52 Porphyromoi	1.50988486 Streptoc 1.07538562 Cutibact	occu 1.06452314 Veillon eriur 0.6843363 Porphy	
	154247 Granulicatell 2.1724962 Novosphingo 1.651097 830328 Caenispirilur 1.84662177 Streptococcu 1.292635	11 Actinomyces 0.4562242 Haemophilus 0.1846 24 Neisseria mu 0.39104932 Porphyromoi 0.0760	66218 03737	Leptotrichia 2.118183 Fusobacteriu 1.694543	79 Rothia dento 1.651 703 Haemophilus 1.618	09711 Streptococcu 50967 Granulicatell	1.83575929 Streptococcu 0.92331088 Rothia mucili 0.90158592 Streptococcu	0.99934825 Capnoc 0.90158592 Granuli	ytoph 0.36932435 Strept atelli 0.34759939 Abjotr	coccu 0.97762329 Por pohia c 0.77123615 Can	phyromo: 1.54247.	23 Rothia mucili (93 Allioprevotell)	0.24983706 Streptoc 0.08689985 Veillone	occu 0.4562242 Granul lla di: 0.33673691 Neisse	
revotella m 1.651 Granulicatell 1.618	543341 Actinomyces 1.12969802 Lautropia mii 1.281772 109711 Lautropia mii 0.76037367 Chryseobacte 1.227460 850967 Kingella orali 0.47794916 Streptococcu 1.173147	85 Streptococcu 0.33673691 Chryseobacti 0.0651 95 Granulicatelli 0.2281121 Granulicatelli 0.054	17489	Oribacterium 0.543124	105 Streptococcu 0.706	06126 Haemophilus	0.89072344 Chryseobacti 0.23897458 Streptococcu 0.19552466 Streptococcu				nella mor 0.738648	71 Neisseria ora i	0.03258744 Campylo	ibact 0.19552466 Neisse	
usobacteriu -16. revotella or -10.6 orynebacter -4.71	58892 Streptococcu 5.938871 Neisseria bac 10.4171 677819 Gemella hae 2.7156202 Leptotrichia 7.09320 143167 Rothia dento 2.4657832 Streptococcu 5.90918	19 Leptotrichia v -23.680209 Neisseria mu -16.36 01 Corynebacter -19.117967 Prevotella or -9.070 97 Selenomona: -9.0918966 Fusobacteriu -7.201	69759 01716 18249	Neisseria elo -18.9984 Capnocytoph -6.64783 Veillonella di -4.90984	179 Veillonella di -17.4 184 Streptococcu -5.82 114 Streptococcu -3.04	34282 Lautropia mii 22898 Prevotella or 14947 Fusobarteriu	-13.969151 Streptococcu -13.382577 Neisseria per	-22.63741 Strepto -2.2268086 Neisser		ella di -18.487943 Veil coccu -11.38388 Nei lobact -4.6056919 Stre	onella di -17.6406 seria ora -4.32326 otococcu -4.19291	69 Streptococcu 74 Lautropia mi 77 Neisseria mu	-5.2465783 Fusobac -2.3897458 Capnocy -2.1616337 Eikenella	toph -30.469259 Strepts	
elenomona: -3.95 ctinomyces -3.31	539431 Neisseria mu -0.9450358 Capnocytoph -5.04019 130567 Streptococcu -0.4996741 Fusobacteriu -4.99574	12 Corynebacter 5.2465783 Streptococcu 6.680 13 Leptotrichia 3.3782316 Capnocytoph 4.670	04258 08668	Neisseria ora -3.50858 Granulicatelli -2.37888	814 Neisseria mu -2.41 833 Prevotella hir -1.72	14708 Anaeroglobu 71345 Dialister invis					ptococcu -3.37823 ptococcu -1.444	16 Streptococcu 71 Streptococcu	-1.0645231 Aggrega -0.6408864 Parvimo	tibad -2.2268086 Neisse nas r -1.9009342 Neisse	
ribacterium -2.91 revotella lo: -2.20	111449 Corynebacte -0.2281121 Campylobact -2.845 050836 Haemophilus -0.1520747 Fusobacteriu -2.70475	97 Campylobact -3.0849446 Streptococcu -3.877 78 Actinomyces -3.0306322 Actinomyces -3.11	79057 17532	Capnocytoph -1.4555 Granulicatell: -0.60829 Leptotrichia : -0.57571	89 Lentotrichia v .0 86	89985 Atonobium ri	-2.84597 Streptococcu -2.4657832 Lautropia mi -1.2709103 Prevotella m	.0 1737997 Strento	a cini -1.2600478 Coryni coccu -0.5213991 Gemel coccu -0.3367369 Porphy	a mor +0 3041495 Gra	nulicatell .1 14056	05 Cannocytoph	.0 5322616 (Fuhacto	rium -1 6510971 Prevot	
tinomyces -2.07 lenomona: -2.07 L: TP3-TP2 (308: t	747339 Streptococcu -0.1520747 Kingella orali: -2.36802 747339 Rothia mucil: -0.1303498 Corynebacter -2.10732	09 Prevotella ou -3.0089072 Capnocytoph -2.661 13 Veillonella di -2.6721703 Streptococcu -2.661	13078 13078	Rothia dento -0.52135 Qipengyuani: -0.40191	991 Veillonella at -0.65 118 Megasphaer: -0.5	17489 Prevotella his 86574 Campylobact	-1.2709103 Prevotella m -1.1731479 Haemophilus	-0.1412123 Rothia -0.1086248 Granuli	dento -0.3367369 Actino catelli -0.3041495 Kingel	oaculu -0.271562 Stre a orali: -0.1846622 Roti	ptococcu -0.74951 nia dento -0.74951	12 Granulicatell 12 Granulicatell	-0.2498371 Trepone -0.2389746 Brachym	ma s -0.9341734 Atopol iona: -0.5974365 Prevot	
cies_nam diff_vo votella or 7.343	alue species nam diff value 303715 Streptococcu 61,9487291 816424 Gemella haei 2.67217032	species_nam diff_value Leptotrichia v 19.400391	species_nam diff_value Streptococcu 22,930697	species_nam.diff_value 4 Neisseria elor 18.94416	species_nam diff_v 668 Streptococcu 16.52 736 Veillonella dr 13.73	18336 Lautropia mii	diff_value species_name 13.9148381 Streptococcu	10.4822942 Strepto	nam diff_value specie coccu 4.88811645 Veillor la per 2.60699544 Granu	ella di 32.6417554 Stre	ptococcu 19.08537	91 Streptococcu	4.07343037 Fusobac	nam diff_value specie: teriu 51.1079731 Strepto	
sobacteriu 6.148 rynebacte 4.594 tinomyces 3.226	482946 Rothia dento 2.57440799 615685 Streptococcu 1.30349772	Corynebacter 18.9441668 Selenomonar 8.88550945 Corynebacter 5.10536607	Streptococcu 10.449706 Veillonella di 8.2772105 Streptococcu 4.5513795	7.94047 11 Capnocytoph 6.615250 14 Streptococcu 4.638279	92 Neisseria mu 7.234 38 Streptococcu 2.726	36259 Prevotella or 41234 Fusobacteriu 48273 Anaeroglobu	11.0905931 Streptococcu 10.3845318 Neisseria per	2.08559635 Strepto 0.98848577 Neisser	coccu 2.18335868 Campy la mu 2.04214643 Strept	lobact 4.86639148 Stre coccu 4.55137954 Stre	onella di 9.178796 ptococcu 5.920052 ptococcu 3.019769	14 Lautropia mi 72 Streptococcu	2.23767108 Capnocy 1.70540952 Eikenella 0.87986096 Aggrega	cor 4.55137954 Neisse tibac 2.1724962 Prevot	
lenomona: 3.106	356941 Streptococcu 1.29263524 666956 Streptococcu 0.64088638 578318 Streptococcu 0.59743645	Leptotrichia (3.37823159 Lachnoanaer 2.52009559 Campylobact 2.46578318	Prevotella de 4.2472300 Prevotella m 3.9104931 Prevotella or 1.9335216	2 Cannocytoph 1 422989	102 Granulicatelli 1.575	05974 Solobacterius 68607 Atonobium ri	3.93221812 Fusobacteriu 2.84597002 Granulicatell	0.6300239 Neisser	coccu 1.29263524 Strept la cini 1.19487291 Coryni coccu 1.0862481 Gemel	bacter 0.4562242 Stre a mor 0.30414947 Nei-	ptococcu 2.465783	18 Haemophilus	0.48881164 Johnson 0.4562242 (Fuhacto		
tinomyces 1.94	214643 Rothia mucil: 0.55398653 443841 Streptococcu 0.39104932 748425 Feeerthia cat 0.31501195	Actinomyces 2.44405822 Prevotella ou 2.30284597 Veilignella di 2.20508364	Atopobium r 1.6076471 Streptococcu 1.5967847	9 Neisseria ora 1.205735 1 Rothia dento 0.879860	39 Neisseria bac 0.68 96 Leptotrichia v 0.608	43363 Prevotella los 29894 Prevotella his 67412 Prevotella mi	2.33543341 Porphyromoi 1.27091028 Caenispirillur 1.10797306 Gemella haei	0.35846187 Rothia (0.24983706 Strepto	mucili 0.58657397 Actino coccu 0.55398653 Neisse	naculu 0.27156202 Abii na mu 0.24983706 Roti	trophia (1.737996) nia dento 1.661959	96 Streptococcu 59 Capnocytoph	0.4019118 Campylo 0.34759939 Trepone	mas 0.9124484 Strepts	
bsiella pri obacteriu -6.53 isseria ora -6.32	353465 Veillonella di +13.502064 392136 Neisseria mu +10.460569 219639 Europacteriu -6.5609385	Alloprevotell -22.050836 Leptotrichia : -11.459917	Klebsiella pn -22.80034 Fusobacteriu -9.493808 Prevotella pli -8.255485	Streptococcu -29.1657	61 Streptococcu -15	58766 Streptococcu	-48.099066 Rothia mucil: -12.296328 Veillonella di	-5.3334782 Strepto -2.7264827 Rothia	coccu -15.479035 Strept lento -0.4236368 Strept	coccu -36.019987 Can	pylobact -16.4457 otella lo: -7.86443	96 Fusobacteriu 62 Veillonella di	-1.8574842 Veillone -1.6836846 Prevotel	lla di -32.891592 Gemel la mi -10.525744 Neisse	
tropia mii -5.82 obacteriu -4.12	222898 Kingella deni -6.1264393 277428 Klebsiella pn -4.7686292	Magnetovibr -6.1807517 Fusobacteriu -4.5296546 Selenomonar -3.975668	Fusobacteriu -5.920052 Campylobact -4.746904	Prevotella ori -1.47729	Magnetovibr -2.95 185 Fusobacteriu -2.53 174 Gemella haer -1.66	19596 Actinomyces	-3.704106 Rothia dento -2.2050836 Streptococcu	-0.8038236 Abiotro -0.6734738 Granuli	ia mii -0.3801868 Gemel phia c -0.3584619 Rothia catelli -0.3475994 Strept	dento -3.7366935 Lau ecoccu -2.552683 Fusi	seria ora -5.58331 ropia mii -3.29133 sbacteriu -3.03063	17 Porphyromo 22 Porphyromo	-1.1188355 Streptoc -0.6300239 Streptoc -0.4670867 Streptoc	occu -6.1155768 Neisse occu -3.3890941 Streoti	
totrichia -1.77	705844 Abiotrophia -3.8235933 705844 Prevotella m -3.725831 316098 Leptotrichia : -2.84597	Treponema n - 3.2370193 Prevotella lot - 3.0849446 Caenispirillur - 2.6721703	Paludibacter -2.444058 Fusobacteriu -2.335433	4 Parvimonas r -1.27091 2 Streptococcu -1.1948 4 Gemella haer -0.7603	103 Prevotella de -1.36 129 Prevotella hir -1.26 137 Fusobacteriu -1.03	86726 Gemelia mor 00478 Veillonella di 19357 Streptococcu	-1.2709103 Streptococcu -1.2274604 Campylobact -1.1405605 Leptotrichia	-0.586574 Leptotr -0.5757115 Fusoba -0.5105366 Eggerth	chia I -0.3258744 Neisse teriu -0.3150119 Rothia ia cat -0.1846622 Lautro	na per -0.3584619 Lep mucil: -0.3150119 Cor pia mii -0.2281121 Lep	nebacte -1.59678 netrichia : -1.50988 otrichia : -1.36867	47 Prevotella lo 49 Alloprevotell 26 Alloprevotell	-0.4453617 Leptotrii -0.4453617 Leptotrii -0.4344992 Atopobii	thia i -3.2370193 Lautro thia i -3.2044319 Chryse um p -2.1942212 Rothia	
npylobact -1.33	360852 Lautropia mi -2.7807951 948729 Capnocytoph -2.1507712 t1-t0)	Prevotella ve -2.4006083 Selenomonar -1.987834	Capnocytoph -1.966109	Alloprevotell -0.59743	65 Caenispirillur -1.03	19357 Streptococcu	-1.0753856 Stomatobacu -0.564849 Chryseobacti	-0.4996741 Actinon	tyces +0.1737997 Strept	coccu -0.1194873 Cor	mebacte: -1.24918	53 Prevotella m	-0.293287 [Clostrid	ium] -2.0530089 Porphy	
cies_nam.diff_vi phyromor 13.74	alue species_nam diff_value 410385 Streptococcu 30.2194221 164675 Streptococcu 3.12839453	species_nam diff_value Eggerthia cat 8.75515968		species_nam.diff_value Streptococcu 27.80795	species_nam.diff_v streptococcu 46.45	14186 Streptococcu	diff_value species_name 30.7190962 Gemella hae 5.70280252 Granulicatell		Abiotr	ophia o 15.1857484 Por	cies_nam diff_value phyromot 10.76471	species_nam c	21.4208125 Streptoc	nam diff_value specie: occu 63.4912014 Neisse	
sseria mu 9.02 phyromoi 7.060	267217 Actinomyces 2.60699544 061264 Fusobacteriu 1.05366066	Fusobacteriu 8.61394743 Leptotrichia : 5.44210298 Klebsiella pni 2.47664567		Veillonella ra 5.65935 Fusobacteriu 3.432543 Neisseria cini 2.118183	326 Alloprevotell 6.951 199 Klebsiella pnr 4.11 179 Streptococcu 4.073		3.7692809 Abiotrophia (3.57375625 Veillonella ra	2.23767108 0.99934825	Strept Rothia Neisse	dento 6.09385184 Cap la elo 4.47534217 Fusi	ptococcu 8.842059 nocytoph 6.767325 obacteriu 6.332826	42 Parvimonas r	15.7940474 Alloprev 1.62937215 Gemella 0.858136 Veillone	haer 7.60373669 Porphy	
totrichia: 3.497	445579 Veillonella ra 0.4562242 771888 Porphyromo: 0.31501195 907017 Granulicatell: 0.27156202	Paludibacter 2.16163372 Capnocytoph 2.08559635 Streetococcu 2.06387139		Granulicatelli 1.303497 Rothia dento 0.868998	772 Gemella mor 1.292 848 Porphyromoi 1.184	63524 Veillonella di 01043 Streptococcu	2.20508364 Streptococcu 1.67282207 Streptococcu 1.40126005 Granulicatell	0.94503585 0.70606126	Aggrey Street	coccu 1.42298501 Egg	nella mor 2.096458 erthia cat 1.651097	B3 Prevotella lo	0.54312405 Chryseol 0.53226157 Streptoc	bact 1.06452314 Actino bact 0.98848577 Porphy occu 0.28242451 Allopre	
obacteriu 1.412	042146 Neisseria mu 0.26069954 212253 Haemophilus 0.2281121 401043 Streptococcu 0.16293721	Neisseria elo: 1.73799696 Leptotrichia · 1.46643493 Tannerella fo 1.40126005		Haemophilus 0.771236	515 Fusobacteriu 0.8	58136 Fusobacteriu	1.24918531 Novosphingo 1.20573539 Micrococcus 1.1405605 Klebsiella pn	0.33673691	Actino	nyces 0.51053661 Con	mebacter 1.357810	12 Actinomyces	0.48881164 Actinom	hilus 0.1737997 Capno yces 0.08689985 Aggreg iculu 0.08689985 Granul	
eptococcu -6.18 inomyces -4.54	807517 Veillonella di -19.758853 405171 Streptococcu -7.6363241	Magnetovibr -14.436237 Actinomyces -5.9526396		Sphingomona -5.60504 Caenispirillur -3.36738	Leptotrichia: -13.7	08451 Lautropia mii 48077 Microlunatus	-47.349555 Streptococcu -3.5194438 Haemophilus	-12.404953 -2.1399088	Strept Strept	coccu -19.096242 Veil	ropia mi -18.7269	17 Streptococcu 17 Streptococcu	-23.408647 Veillone -5.9417771 Leptotrii	lla di -19.356941 Strepti chia -9.6132957 Lautro	
llonella di -4.37 inerella fo -3.66 ingomon: -3.18	775798 Streptococcu 6.6152509 606561 Streptococcu -0.6843363 827069 Prevotella m -0.4888116	Veillonella di -5.7897024 Leptotrichia : -5.3769281 Prevotella de -4.7034543		Veillonella de -3.09580 Lautropia mir -2.98718 Streptococcu -2.76993	23 Capnocytoph -7.12 27 Fusobacteriu -6.10	57875 Cardiobacter 47143 Leptotrichia (-1.1948729 Alloprevotell -0.7277862 Veillonella di -0.6734738 Streptococcu	-0.3041495 -0.1412123	Gemel Granu	lobact -2.3354334 Stre a haer -1.8248968 Pre- icatell: -1.6293721 Gra	otella los -6.21333 nulicatell -2.82424	91 Rothia dento 51 Haemophilus	-2.281121 Caenispi -2.1942212 Prevotel -1.9769715 Streptoc	la m -5.3117532 Prevot occu -4.5296546 Actino	
00m0021 -2 94	501195 Selenomona: -0.3801868 437323 Capnocytoph -0.271562 568325 Actinomyces -0.271562	Prevotella hir -3.8561808 Alloprevotell -1.422985 Olsenella uli -1.4121225		Fusobacteriu -2.60699 Granulicatell: -2.5744 Prevotella loc -2.35719	954 Caenispirillur 4.03 108 Lautropia mir -2.03 1584 Actinomyces -1.90	34304 Actinobaculu 12839 Prevotella los 09342 Tessaracoccu	-0.4996741 Streptococcu -0.4996741 Porphyromoi -0.4562242 Actinomyces	-0.0760374 -0.0760374 -0.0651749						a mu -3.4434065 Kingell ionas -2.9545948 Actino um p -2.2919835 Neisse	
otrichia -2.81	133826 Atopobium r -0.1955247 137085 Rothia dento -0.1955247	Caenispirillur -1.3578101 Megasphaer; -1.2709103		Qipengyuani: -1.9878	34 Neisseria mu -1.46	64349 Rothia dento	-0.3367369 Porphyromo -0.2063871 Haemophilus	-0.0543124	Coryne	bacter -0.3693244 Stre	ptococcu -0.72778	62 Alloprevotell	-0.8146861 Selenom	onas -2.009559 Pseudo onas -1.7814469 Gemel	
ies_nam diff_vi	value species_nam.diff_value species_nam.diff_value 403433 Veillonella di 28.6660873 Streptococcu 16.0112	species_nam diff_value 97 Magnetovibr 11.7314795	species_nam diff_value Neisseria elo 9.8522702	species_nam.diff_value	71 Prevotella loc 16.40	alue species_nam 23463 Lautropia mii	diff_value species_name 62.6439279 Granulicatell 3.28046926 Alloprevotell	liff_value 5.62676515	Strept	_nam.diff_value spe rcoccu <mark> 32.4245058</mark> Veil	lonella di 10.22159	46 Streptococcu	30.6973713 Veillone	nam diff_value specie: Ila di <mark> 11.3838801</mark> Lautro	
onella di 7.147 ptococcu 6.148 nomyces 4.529	751249 Streptococcu 13.6541386 Caenispirilluf 14.5883 816424 Streptococcu 5.19226591 Actinomyces 12.25287 965457 Actinomyces 1.27091028 Eikenella.com 8.97240	12 Actinomyces 5.74625244 86 Prevotella de 4.63827938 93 Fusobacteriu 3.81273083	Streptococcu 7.484249 Streptococcu 7.2452748 Pseudopropi 6.2133391	4 Alloprevotell 5.04019: 12 Lautropia mir 4.8012: 13 Sphingomon: 4.594829	118 Leptotrichia : 10.8 166 Fusobacteriu 7.94 146 Capnocytoph 4.562	04736 Tessaracoccu	3.20443189 Streptococcu 2.34629589 Granulicatell	1.55333478	Rothia	lobact 1.62937215 Pre- dento 1.3686726 Fusi	obacteriu 2.911144	48 Streptococcu	3.4868564 Leptotrii 2.78079513 Caenispi 2.21594612 Neisserii	rillur 5.39865305 Prevot	
erella fo 3.671 ngomon: 3.334	478166 Selenomona: 0.38018683 Actinomyces 2.520095	07 Prevotella hir 2.95459483 59 Leptotrichia i 2.75907017	Streptococcu 5.3226156 Lautropia mi 3.6715185 Klebsiella pp. 3.6497936	7 Neisseria elo: 1.249185		58288 Capnocytoph	1.93352162 Haemophilus 1.27091028 Veillonella ra	0.47794916	Berger	icatell: 0.98848577 Nel: ella zo 0.87986096 Gra	nulicatell 2.183358	68 Gemella haer	1.90093417 Atopobii	onas 2.66130784 Actino um p 2.04214643 Kingell ionas 1.79230936 Actino	
otrichia (2.845	666956 Prevotella m 0.35846187 Actinomyces 2.487508 459483 Streptococcu 0.33673691 Lautropia mir 2.085596 597002 Capnocytoph 0.23897458 Corynebacter 1.824896	81 Selenomona: 0.84727352	Fusobacteriu 1.2491853	1 Porphyromoi 0.966760	81 Capnocytoph 1.509	88486 Leptotrichia	0.94503585 Porphyromoi 0.74951119 Actinomyces 0.64088638 Haemophilus	0.0543124	Gemei Strept	a haer 0.6300239 Nei rcoccu 0.4562242 Stre	seria mu 2.020421 ptococcu 1.575059	46 Haemophilus 74 Alloprevotell	0.69519878 Selenom 0.58657397 Selenom	ionas 1.59678471 Prevot ionas 1.30349772 Leptot	
ptococcu -5.85	233326 Atopobium r 0.19552466 Actinomyces 1.661959 0.02998 Streptococcu 28.742125 Veillonella di 44.481 548773 Streptococcu 24.136433 Streptococcu 5.34434	59 Porphyromoi 0.81468607 86 Caenispirillur 23.169672 06 Leptotrichia 4.3667174	Fusobacteriu -18.3901 Alloprevotell -7.875298	9 Granulicatelli 0.966760 8 Prevotella lot -7.81013 7 Eggerthia cat -6.5935	38 Veilionella di -39	75668 Streptococcu 59983 Fusobacteriu	0.48881164 Prevotella m -\$4.844667 Streptococcu -6.7456007 Veillonella di	-5.9417771	Kingel Strept Lautro	coccu -11.861829 Stre	nella mor 1.422985 ptococcu -12.4592 ropia mir -10.3410	01 Porphyromo: 66 Neisseria per 82 Fusobacteriu	-9.6458831 Streptoc -7.7666739 Streptoc	occu -14.077775 Rothia	
pylobact -5.04 otococcu -4.61 seria mu -4.21	401912 Rothia dento -0.7495112 Fusobacteriu -4.05170 165544 Gemelia haer -0.2498371 Alloprevotelt -3.71496 146426 Actinomyces -0.1412123 Streptococcu -3.62806	54 Alloprevotell -3.9322181 85 Prevotella loi -1.9552466 87 Klebsiella pni -1.3252227	Leptotrichia: -2.672170	7 Caenispirillur -3.35650 13 Leptotrichia : -3.23701	956 Streptococcu -7.67	97741 Streptococcu 51792 Streptococcu	-4.8446665 Streptococcu -3.2044319 Gemella haei -1.6619596 Streptococcu	-0.7495112	Neisse Strept Leptot	ria mu -5.5398653 Cap rcoccu -4.8446665 Acti	nocytoph .7 28872.	27 Paludibacter	.6 2676515 Strenton	occu -1.6185097 Campy la de -0.4344992 Strepte thia : -0.3258744 Neisse	
ohyromoi -3.03 nulicatell -1.94	306322 Streptococcu -0.1194873 Neisseria mu -2.98718 443841 Neisseria mu -0.0760374 Streptococcu -2.35715	23 Prevotella ou -1.1405605 84 Prevotella sa -0.9667608	Granulicatell -1.727134 Campylobact -1.683684	IS Qipengyuani: -1.83579 IG Caenispirillur -1.19487	93 Porphyromoi -1 729 Streptococcu -0.86	40126 Gemelia mor 89985 Streptococcu	-1.2057354 Rothia mucili -0.9450358 Streptococcu	-0.4996741 -0.4670867	Eggert Neisse	nia cat -2.5635455 Cap ria per -2.2376711 Acti	nocytoph -2.68303 nomyces -1.62937	28 Klebsiella pn 21 Fusobacteriu	-2.0964588 Granulic -1.1622855 Magneto	atelli -0.3041495 Porphy ovibri -0.2824245 Coryne	
phyromo: -1.27 nella hae: -1.08	967847 Streptococcu - 0.0760374 Actinomyces - 1.32522 709103 Campylobact - 0.0543124 Actinomyces - 1.03193 862481 Actinomyces - 0.0434499 Prevotella mi - 0.94503	57 Prevotella ve -0.6300239 58 Veillonella at -0.5974365	Oscillibacter -1.184010 Paludibacter -0.945035	2 Aggregatibac -1.14056 4 Magnetovibr -1.06452 8 Veillonella dt -0.78209	131 Leptotrichia I -0.2 186 Neisseria elo: -0.2	93287 Veillonella di 81121 Parvimonas r	-0.8472735 Streptococcu -0.8146861 Neisseria per -0.6843363 Neisseria elo	-0.1520747 -0.1520747	Abiotr Rothia	ella di -1.9769715 Kot ophia c -1.9226591 Hae mucil: -1.9009342 Qip	mophilus -0.40191 engyuani -0.36932	18 Alloprevotell 44 Neisseria elo	-0.9015859 Prevotel -0.9015859 OTU_23	occu -0.271562 Neisse la sal -0.2172496 Chryse 3 -0.1955247 Bergey	
TP2-TP1 (218: t cies_nam diff_v: phyromo: 19.96	value species_nam diff_value	species_nam.diff_value Streptococcu_87_2474473		species_nam.diff_value Streptococcu_20.42146	species_nam.diff_v 43 Streptococcu 21.51	alue species_nam 85748 Streptococcu	diff_value species_name	7.85357376 Strepto			cies_nam diff_value	species_nam c	8.73343472 Actinoba	culu 52.574408 Veillon	
seria mu 17.39 Ionella di 16.23	908321 Actinomyces 1.55333478 394091 Granulicatell 1.03193569 803824 Abiotrophia 1.0.20538714	Streptococcu 0.20638714 Streptococcu 0.13034977 Novosphingo 0.11948729		Fusobacteriu 4.149467 Leptotrichia 1 3.573756	774 Neisseria mu 9.185 525 Streptococcu 7.625	65892 Streptococcu 46166 Haemophilus 70692 Wallonalla di	6.26765153 Gemelia hae 2.15077124 Haemophilus 0.69519878 Streetersory	4.61655442 Granuli 1.90093417 Neisser	a ner fi 73864871 Canno	ria ora 23.6041712 Ger pia mir 3.04149468 Stre cytoph 2.98718227 Veil	Ionella di 5 333478	48 Streptococcu	5.74625244 Streptoc 3.43254399 Gemella 1.9996965 Arthorn	haer 4 69259179 Poroby	
ptococcu 9.265 ptococcu 4.779 strophia 3.48	569629 Capnocytoph 0.14121225 949164 Lautropia mi 0.10862481 868564 Actinomyces 0.08689985	Rothia mucil: 0.10862481 Granulicatell: 0.08689985 Campylobact 0.0543124		Neisseria mu 1.54247	23 Streptococcu 2.368	02086 Granulicatell	0.69519878 Streptococcu 0.51053661 Haemophilus 0.46708668 Rothia mucili 0.27156202 Granulicatell	0.6843363 Granuli	atelli 0.09776233 Abiotr	ohia c 0.858136 Gra	nulicatell 1.390397	57 Veillonella di 1	0.71692375 Chryseol	bacts 0.35846187 Porphy	
ptococcu 2.378 moanaer 1.151	888334 Fusobacteriu 0.08689985 142299 Actinomyces 0.04344992	Rothia dento 0.0543124 Neisseria cini 0.04344992		Corynebacter 1.292635 Streptococcu 1.227460	324 Haemophilus 1.803 335 Streptococcu 1.733	17184 Haemophilus 99696 Porphyromor	0.07603737 Chryseobacti 0.0543124 Streptococcu	0.53226157 Haemo 0.41277428 Coryne	ohilus 0.06517489 Tessar oacter 0.0543124 Strept	coccu 0.61916142 Fusi coccu 0.53226157 Ger	obacteriu 1.129698 nella mor 1.010210	73 Streptococcu	0.33673691 Pseudop 0.31501195 Caenispi	ropii 0.24983706 Veillon rillur 0.16293721 Neisse	
obacteriu -39.7 nispirillur -12.7	538562 Bergeyella zc 0.03258744 702368 Streptococcu 21.312188 752553 Streptococcu 16.054747	Paracoccus m 0.04344992 Caenispirillur 37.725896 Magnetovibr -3.3999566		Streptococcu -15.3160 Fusobacteriu -7.85357	98 Veillonella di -23.5 38 Streptococcu -17	62633 Lautropia mii 29307 Rothia dento	-3.5194438 Veillonella ra	-18.466218 Neisser -0.9124484 Strepto	a mu -7.2778623 Strept	10.395394 Stre	ptococcu -33.5759 ptococcu -1.22746	29 Streptococcu 04 Streptococcu	-7.7775364 Veillone -6.5066261 Strepton	lla di -38.464045 Strepti occu -6.3654139 Gemel	
otella pa -7.26 otella or -5.33	669998 Streptococcu -5.0945036 334782 Actinomyces -3.9539431 587443 Streptococcu -2.9220074	Klebsiella pni -2.9545948 Leptotrichia v -2.84597 Veillonella di -2.5852705					-3.4434065 Alioprevotell -2.552683 Haemophilus -2.0204215 Porphyromo		romoi -4.2363676 Strept coccu -4.0842929 Kingel coccu -3.8670432 Campi	coccu -4.6165544 Stre a orali: -2.7482077 Can lobact -2.3462959 Rot	ptococcu -1.21659 pylobact -0.52139 nia dento -0.47794	79 Granulicatell 91 Lautropia mi 92 Streptococcu	-2.4766457 Prevotel -1.4121225 Streptoc -1.2165979 Leptotrii	la sa -5.8005648 Lautro occu -4.4970671 Neisse thia -3.117532 Strepte	
pylobact -2.82 nomyces -2.07	242451 Actinomyces -1.3795351 747339 Streptococcu -0.7712362 338475 Haemonbilus -0.543174	Actinobaculu -2.4657832 Selenomona: -1.9986965 Leptotrichia: -1.9117967		Actinomyces -1.81403	Prevotella ou -1.01 43 Fusobacteriu -0.80	02107 Neisseria mu 38236 Gemella haer	-2.009559 Neisseria elo -1.8792092 Neisseria mu -1.4881599 Neisseria cin	-0.0325874 Bergeyi -0.021725 Veilloni	illa zo -1.1079731 Porphy illa di: -0.8038236 Actino	romo: -0.7277862 Por nyces -0.7169237 Neb	seria ora -0.08689	48 Rothia dento 98 Streptococcu	-1.1840104 Caenispi -1.0862481 Atopobii	rillur -3.0849446 Strepts um p -2.6287204 Actino	
otrichia : -1.09	731479 Alloprevotell -0.1955247 971106 Rothia dento -0.1520747	Selenomonar -1.8140343 Prevotella de -1.7271345		Gemella haer -1.47729	974 Megasphaer: -0.64	08864 Fusobacteriu	-1.2274604 Prevotella m -1.0319357 Streptococcu	-0.0108625 Strepto	coccu -0.4019118 Neisse	ia mu -0.5539865 Stre	ptococcu -0.04344	99 Granulicatell	-0.4344992 Atopobii	um ri -2.1073213 Cutiba	
obacteriu 38.18	value species_nam diff_value 816207 Streptococcu 12.1008038	species_nam.diff_value Caenispirillur_26.2111666		species_nam.diff_value Veillonella de 10.94938	08 Veillonella di 25.60	alue species_nam 28677 Lautropia mir	diff_value species_name 29.6219857 Streptococcu 4.00825549 Granulicatell	liff_value species 5.9417771 Strepto	nam diff_value specie toccu 13.8713882 Strept la mu 7.13665001 Strept	_nam diff_value spe coccu <mark> 50.5322616</mark> Stre	cies_nam.diff_value ptococcu <mark> 59.64588</mark>	species_nam c	diff_value species_ 5.78970237 Veillone 3.23701934 Prevotel	nam diff_value specie:	
ispirillur 8.668 otella pa 5.43 otella or 3.008	825983 Streptococcu 11.3187052 312405 Actinomyces 2.2268086 890723 Streptococcu 1.85748425	Streptococcu 4.82294156 Magnetovibr 2.46578318 Actinobaculu 2.44405822		Fusobacteriu 7.853573 Leptotrichia (3.508581 Prevotella los 3.237019	376 Streptococcu 19.30 136 Streptococcu 17.35 134 Streptococcu 3.791	82446 Rothia dento	4.00825549 Granulicatell 3.55203128 Haemophilus 2.19422116 Veillonella ra	2.61785792 Strepto	coccu 4.92070389 Granu	coccu 13.0892896 Stre icatell: 2.83510754 Ger lobact 2.79165761 Gra	nella haer 1.575059	74 Granulicatell	2.30284597 Caenispi	rillur 2.98718227 Lautro	
iella nor 3 008	890723 Veillonella di 1.82489681 287204 Actinomyces 0.93417337 781012 Haemophilus 0.58657397	Klebsiella pni 2.2268086 Prevotella de 1.06452314 Selenomona: 0.89072344		Prevotella mi 2.183358 Actinomyces 1.922659	68 Streptococcu 3.475 914 Klebsiella pnr 3.421	99392 Leptotrichia I 68151 Campylobact	1.5424723 Alloprevotell 1.28177276 Streptococcu 1.04279818 Granulicatell	1 64023463 Pornhy	omoi 1.41212253 Kingel	orrali 2 19422116 Gra	nulicatell 0 249837	N6 Haemonhilus	1 35781012 Atonobii	um n 2 41147078 Neisse	
bacteriu 1.118 iocytoph 1.064	883554 Alloprevotell 0.19552466 452314 Prevotella int 0.10862481	Gemella haei 0.82554856 Prevotella pli 0.72778623		Neisseria ora 0.836411 Campylobact 0.564849	104 Neisseria mu 0.879 101 Prevotella ou 0.879	86096 Leptotrichia (86096 Corynebacter	0.74951119 Haemophilus 0.71692375 Abiotrophia i	0.11948729 Veilloni 0.07603737 Fusoba	illa di: 0.60829894 Berge _l :teriu 0.46708668 Tessar	ella zo 0.64088638 Stre acoccu 0.34759939 Orit	ptococcu 0.021724 acterium 0.021724	96 Gemella mor i 96 Granulicatell i	0.43449924 Prevotel 0.36932435 Stomato	la ou 1.64023463 Cutiba bacu 1.24918531 Actino	
otella de -41.0 obium ri -4.97	124484 Caenispirillur 0.07603737 105866 Streptococcu <mark> 27.628289</mark> 750163 Granulicatell -0.8907234	Prevotella hir 0.72778623 Leptotrichia 12.643928 Leptotrichia 12.643928		Alloprevotell -11.5685 Neisseria mu -5.9852	42 Capnocytoph -25.0 27 Fusobacteriu -11.4	59744 Streptococcu 59917 Streptococcu	.13 121877 Gemella haer	-1.9335216 Granuli	catell -18.335868 Strept	coccu -50.847274 Rot	nia dento -15.6854	Neisseria per	-2.84597 Streptoc	occu -18.82468 Strepts	
nomonar -3.92 ster invis -2.99 otella hir -2.5	213556 Streptococcu -0.8472735 980448 Fusobacteriu -0.5757115 552683 Campylobact -0.3584619	Veillonella di -10.058657 Prevotella loc -2.5635455 Lachnoanaer -1.6402346		Streptococcu -3.68238 Streptococcu -2.845 Streptococcu -1.61850	811 Leptotrichia : -10 97 Kingella orali -6.48 97 Streptococcu -4.80	15642 Fusobacteriu 49012 Streptococcu 12166 Neisseria mu	-4.8012166 Rothia mucil: -2.6830328 Neisseria per -2.1833587 Veillonella di	-1.44471 Strepto -1.3360852 Veillone -1.1948729 Gemell	roccu -4.7350417 Abiotr ella ra -4.3667174 Actino n haer -2.4875081 Actino		ropia mii -3.32391 obacteriu -2.5744	Veillonella di Porphyromo	-1.5533348 Streptoc	occu -10.688681 Strepts occu -1.44471 Porphy	
nella di -2.19 eria mu -1.70	942212 Veillonella ra - 0.2498371 054095 Neisseria per - 0.1955247 926352 Streptococcu - 0.1737997	Fusobacteriu -1.6185097 Capnocytoph -1.4664349 Prevotella mr -1.2600478		Gemella haer -1.06452	31 Capnocytoph -2.37	88833 Streptococcu	-2.1833587 Veillonella di -1.6728221 Chryseobacti -1.5750597 Lautropia mi -1.2383228 Streptococcu	-0.8689985 Strepto	coccu -0.3801868 Veillor	ella di -0.7820986 Egg	erthia cat -1.62937.	21 Porphyromox	-0.7603737 Megaspl	haer: -0.3367369 Prevot	
ngyuani -1.28	817728 Alloprevotell -0.1737997 491853 Gemella haer -0.1520747	Actinomyces -1.1188355 Prevotella sa -1.0536607		Fusobacteriu -0.68433	663 Campylobact -1.53	16098 Gemella mor	-0.9233109 Rothia dento -0.7386487 Streptococcu	-0.6082989 Granuli	atelli -0.1086248 Coryni	bacter -0.586574 Acti	nomyces -1.46643	49 Clavibacter n	-0.3367369 Prevotel	la pa -0.1520747 Chryse	
atobacu -1.24				species nam-diff value	species_nam.diff_v	alue_species_nam	diff_valuespecies_name	liff_value	specie	nam diff_value spe coccu <mark> 16.8151206</mark> Por	phyromo: 9.265696.	29 Lautropia mi	12.9372149 Prevotel	nam diff_value specie: la lot 10.3193569 Strepte	
P2-TP1 (21L: t ies_nam.diff_v otrichia : 6.941	value species_nam.diff_value 112535 Streptococcu 6.72387573	species_nam.diff_value Fusobacteriu 20.4649142		Lautropia mil 24.50579	71 Kingella orali 58.16	85857 Streptococcu	17.7492939 Eggerthia cat	32.000/3		bacter 4.44275473 Fusi	obacteriu 6.452313	71 Fusobacteriu	9.4829459 Leptotrii		
P2-TP1 (21L: t es_nam diff_vi otrichia : 6.941 bacteriu 6.484 nebacter 3.410	value species_nam.diff_value 112535 Streptococcu	Fusobacteriu 20.4649142 Capnocytoph 10.0369324 Streeteroccu 9.70019552		Streptococcu 7.462524 Granulicatelli 1.477297	71 Kingella orali 58.16 144 Capnocytoph 7.545 741 Neisseria bac 2.791	85857 Streptococcu 42429 Neisseria elo 65761 Campylobact	17.7492939 Eggerthia cat 10.7755811 Altererythrol 3.89963068 Corynebacte 2.82424506 Porphyromo	12.013904 7.34303715 5.24657832	Rothia	dento 1.98783402 Leo	otrichia : 5.181403 nella con 4.985878	43 Porphyromo	4.76862915 Porphyri 4.23636759 Streptoc	omoi 6.47403867 Allopro occu 6.13730176 Fusoba	
rP2-TP1 (21L: t ies_nam diff_v: otrichia : 6.941 bacteriu 6.484 nebacte: 3.410 otococcu 3.106 ella deni 1.955 iocytoph 1.759	ralue pecies, nam diff value 11555 Sreptococcu 27233577 490113 Streptococcu 5.2574408 5005999 5005999 5005999 5005999 5005999 5005999 5005999 5005999 5005999 5005999 5005999 5005999 5005999 5005999 5005999 5005999 5005999	Fusobacteriu 20.4649142 Capinocytoph 10.0369324 Streptococcu 9.70019552 Leptotrichia 5.509450358 Leptotrichia 4.70345427 Klebsiolia pnd 4.49705713		Streptococcu 7.462524 Granulicatelli 1.477297 Neisseria mu 1.260047 Streptococcu 0.738648 Neisseria per 0.608298	771 Kingella orali: 58.16 144 Capnocytoph 7.545 741 Neisseria bac 2.791 779 Tessaracoccu 2.107 871 Leptotrichia I 1.727 884 Bergeyella zo 1.075	42429 Neisseria elo 65761 Campylobact 32131 Capnocytoph 13448 Leptotrichia I 38562 Leptotrichia:	3.89963068 Corynebacte 2.82424506 Porphyromo 2.02042146 Neisseria elo 1.96610906 Porphyromo	12.013904 7.34303715 5.24657832 5.18140343 4.60569194	Rothia Neisse	dento 1.98783402 Lep la ora 1.83575929 Eike	nella con 4.985878	Porphyromoi Porphyromoi Leptotrichia: Prevotella lo	4.23636759 Streptoc 3.74755594 Fusobac 3.23701934 Neisseria	occu 6.13730176 Fusoba teriu 5.56159027 Porphy a per 4.56224202 Prevot	
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