Original Article

Impact of Perioperative Management on Ocular Microbiota Composition and Diversity: A Study of Intravitreal Injection Patients with 16S rRNA Sequencing

Hamidreza Taherkhani [1], MS; Azadeh Kavianfar [1,*], MS; Mahdieh Salimi [2], PhD

1. Laboratory of Systems Biology and Bioinformatics, Department of Bioinformatics, Kish Int. Campus, University of Tehran, Kish, Iran.

2. Department of Medical Genetics, Institute of Medical Biotechnology, National Institute of Genetic Engineering and Biotechnology (NIGEB), Tehran, Iran

***Corresponding Author:** Azadeh Kavianfar

E-mail: akavianfar@ut.ac.ir

Abstract:

Background: The ocular microbiota, which includes both commensal and pathogenic microorganisms, is constantly exposed to the ocular surface. It has recently become increasingly acknowledged that the ocular microbiota plays a vital role in maintaining eye health and that interventions, including the use of drugs on the surface of the eye, can potentially disrupt the equilibrium of microorganisms within the eye. One area that has received relatively little attention in the literature is the potential effect of these interventions on the microbiota within the vitreous. The aim of this study is to investigate the effect of intravitreal injections on the ocular microbiota of patients, specifically examining changes in the composition and relative abundance of ocular microbes as a result of this treatment.

Material and Methods: In this study, two groups of patients were analyzed. Group A included 19 individuals who had not received intravitreal injections or undergone perioperative management. Group B, on the other hand, consisted of 22 patients who had received one, two, or more two treatments. The microbial samples collected from the ocular surface of these patients were subjected to 16S rRNA sequencing using the HiSeq 2500 platform. Further analysis of the alpha/beta diversity and clustering of operating taxonomic units (OTUs) was carried out.

Results: Our results show a significant difference in beta diversity was observed between group A (15 patients without intravitreal injections or perioperative management) and group B (patients with at least one, twice, or more than twice treatment) with a P value of 0.014. It was found that both the composition and relative abundance of cells were impacted by perioperative management in the lead-up to intravitreal injection. Additionally, a greater diversity of Gram-negative bacteria was observed and the most significant groups of microbiotas were found to be phyla and genera. Conclusion: In conclusion, our study found that perioperative management has a significant impact on the ocular microbiota, altering its composition and disrupting its balance. Therefore, it is important for clinicians to carefully consider perioperative management prior to administering intravitreal injections.

Keywords: Intravitreal Injection; Antimicrobial Resistance; Ocular Surface Microbiota; Perioperative Management.

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Introduction

As populations age in many countries, there will be an uptick in ocular diseases such as age-related macular degeneration (AMD), diabetic retinopathy (DR), polypoidal choroidal vasculopathy (PCV), neovascular glaucoma (NVG), and central and branch retinal vein occlusions (CRVOs and BRVOs). Intravitreal injections (IVTs) are often recommended to treat these diseases [1]. However, there is a significant risk of ocular complications when the ocular surface microbiota is directly injected into the posterior vitreous cavity via intravitreal injections or ophthalmic surgery. Studies have shown that the incidence of endophthalmitis after IVT injections is higher compared to after cataract surgery [2,3]. The ocular surface microbiota is known to be a source of infection in post-procedural endophthalmitis. Despite the use of povidone-iodine and topical antibiotics to sterilize the ocular surface before IVT injections, around 20 % of needles have been found to be contaminated with bacteria [4,5].

Traditionally, the presence of bacteria on used needles was examined through conventional microbiological culture methods and non-selective growth media [6,7]. This approach would often detect gram-positive bacteria such as Staphylococcus epidermidis and Corynebacterium. However, this method can be limited as some bacteria may not be able to grow on standard laboratory media, resulting in an incomplete understanding of the types of bacteria presents.

The discovery of the Human Microbiome Project in 2007 has led to a proliferation of international studies on the bacterial flora of the ocular surface. Traditional methods, such as culture-based techniques, have been used to study the ocular microbiome in healthy individuals, and these studies have generally found few differences in bacterial species. However, with the advent of culture-independent methods, such as 16S ribosomal RNA gene sequencing, a greater level of diversity has been observed in the microbial communities of the ocular surface [8]. It is important to note that this community of microbes is not only limited to the ocular surface, but also interacts with other microbes and organs in the body, influencing the immune response. Further research is needed to fully understand the implications of these findings and their potential impact on ocular health [9] creating an unparalleled reference set of microbiome specimens. To ensure that specimens represented minimally perturbed microbiomes, we first screened potential participants using exclusion criteria based on health history, including the presence of systemic diseases (e.g., hypertension, cancer, or immunodeficiency or autoimmune disorders. The ocular microbiome is a complex and dynamic ecosystem that plays a crucial role in maintaining the health of the ocular surface. As highlighted by Aagaard et al. in 2013, the ocular microbiome is composed of a diverse array of microorganisms [9] creating an unparalleled reference set of microbiome specimens. To ensure that specimens represented minimally perturbed microbiomes, we first screened potential participants using exclusion criteria based on health history, including the presence of systemic diseases (e.g., hypertension, cancer, or immunodeficiency or autoimmune disorders, including both pathogens and mutualists. These microorganisms interact with one another and with the host to regulate various physiological processes, including immunity and inflammation. Recent research has shown that the ocular microbiome is closely linked to a number of ocular diseases, including

glaucoma, macular degeneration, diabetic retinopathy, infectious keratitis, and dry eye [10] dry eye group. For example, studies have found that patients with glaucoma have a different ocular microbiome compared to those without the disease [11]. This suggests that the ocular microbiome may play a role in the development and progression of glaucoma. Given the growing body of evidence linking the ocular microbiome to ocular health, ophthalmologists are becoming increasingly interested in understanding the microbiome of the ocular surface. This includes identifying the specific microorganisms present, as well as understanding how these microorganisms interact with one another and with the host. By understanding the ocular microbiome, ophthalmologists may be able to develop new strategies for preventing and treating ocular diseases [12] because the microbiome, composed of the genetic material of bacteria, fungi, viruses, protozoa, and eukaryotes on the ocular surface, differs from the microbiota, which are the community of microorganisms that colonize the ocular surface. The observed composition of the ocular surface flora depends on harvesting and examining methods, whether with traditional culture or with more refined genetic analysis based on rRNA and DNA sequencing. Environment, diet, sex, and age influence the microbial flora composition, thus complicating the analysis of the baseline status. Moreover, potentially pathogenic organisms can affect its composition, as do various disorders, including chronic inflammation, and therapies applied to the ocular surface. A better understanding of the composition and function of microbial communities at the ocular surface could bring new insights and clarify the epidemiology and pathology of ocular surface dynamics in health and disease. The purpose of this review is to provide an up-to-date overview of knowledge about this topic.",”author”:[{“dropping-particle”:””,”family”:”Aragona”,”given”:”Pasquale”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Baudouin”,”given”:”Christophe”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Benitez del Castillo”,”given”:”Jose M.”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Messmer”,”given”:”Elisabeth”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Barabino”,”given”:”Stefano”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Merayo-Lloves”,”given”:”Jesus”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Brignole-Baudouin”,”given”:”Francoise”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Inferrera”,”given”:”Leandro”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Rolando”,”given”:”Maurizio”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Mencucci”,”given”:”Rita”,”non-dropping-particle”:””,”parse-names”:false,” suffix”:””},{“dropping- particle”:””,” family”: ”Rescigno”,”given”: ”Maria”,”non- dropping- particle”:””,” parse- names”: false,” suffix”:””}, {“dropping-particle”: ””,”family”: ” Bonini”,”given”: ”Stefano”,” non- dropping- particle”: ””,” parse- names”: false,” suffix”: ””},{“dropping-particle”:””,” family”: ” Labetoulle”,” given ”:” Marc”,” non- dropping- particle”: ””,” parse- names”: false, ” suffix”: ””}], ”container-title”: ” Survey of Ophthalmology”,”id”:

”ITEM-1”, ”issue”:”6”,”issued”:{“date-parts”:[[“2021”]]},”page”:”907-925”, ”publisher”:”Elsevier Inc.”,”title”:”The ocular microbiome and microbiota and their effects on ocular surface pathophysiology and disorders”,”type”:”article-journal”,”volume”:”66”},”uris”:[“http://www.mendeley.com/documents/?uuid=960f9e0d-acf1-4037-b3f9-595f79c3af00”]}],”mendeley”:{“formattedCitation”:”(12. Overall, the ocular microbiome is a complex and dynamic ecosystem that plays a critical role in maintaining the health of the ocular surface. Further research is needed to fully understand the interactions between the ocular microbiome and ocular health, but the potential for new treatments is promising [13] the tear proteome was identified using chromatography tandem mass spectrometry. After compositional and functional profiling of the metagenome and functional characterization of the proteome by gene ontology, association studies between the ocular microbiome and tear proteome were assessed. RESULTS: Two hundred twenty-nine taxa were identified with Actinobacteria and Proteobacteria being the most abundant phyla with significantly more Propionibacterium acnes and Staphylococcus epidermidis in lid compared to conjunctival swabs. The lid metagenomes were enriched in genes of the glycolysis lll and adenosine nucleotides de novo and L-isoleucine biosynthesis. Correlations between the phylum Firmicutes and fatty acid metabolism, between the genus Agrobacterium as well as vitamin B1 synthesis and antimicrobial activity, and between biosynthesis of heme, L-arginine, as well as L-citrulline and human vision were detected. CONCLUSIONS: The ocular surface microbiome was found to be associated with the tear proteome with a role in human immune defense. This study has a potential impact on the development of treatment strategies for ocular surface-associated diseases.”,”author”:[{“dropping-particle”:””,”family”:”Zysset-Burri”,”given”:”Denise C”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Schlegel”,”given”:”Irina”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Lincke”,”given”:”Joel-Benjamin”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Jaggi”,”given”:”Damian”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Keller”,”given”:”Irene”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Heller”,”given”:”Manfred”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Lagache”,”given”:”Sophie Braga”, ”non- dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Wolf”,”given”:”Sebastian”,”non-dropping-particle”:””,”parse-names”:false,”suffix”:””},{“dropping-particle”:””,”family”:”Zinkernagel”,”given”: ”Martin S”, ”non- dropping-particle”: ””,”parse- names”: false,”suffix”: ””}],”container-title”:”Investigative ophthalmology & visual science”,”id”:”ITEM-1”,”issue”:”10”,”issued”: {“date-parts”:[[“2021”,”8”]]},”language”: ”eng”, ”page”: ”8”,”publisher- place”: ”United States”,”title”: ”Understanding the Interactions Between the Ocular Surface Microbiome and the Tear Proteome.”,”type”: ”article-journal”,”volume”:”62”},”uris”:[“http://www.mendeley.com/documents/?uuid=41aef4d6-64a9-48f7-a8f8-34bdda252bb5”]}],”mendeley”:{“formattedCitation”:”(13. The use of intravitreal injections (IVTs) has become a common method for

treating ocular diseases such as age-related macular degeneration (AMD) and diabetic retinopathy (DR). However, recent studies have revealed that IVT needles are often contaminated with a wide variety of bacteria, which can pose a risk for complications such as endophthalmitis. This risk is further compounded by the fact that the diversity of bacteria found on the ocular surface and in the conjunctival tissue is similar to that found on the IVT needles. In order to gain a better understanding of this problem, the current study used 16S rRNA gene sequencing data to analyze the levels of contamination on IVT needles. This research provides important insights into the potential risks associated with IVT procedures and highlights the need for improved sterilization methods to minimize the risk of complications [4].

The ocular surface microbiome plays a crucial role in maintaining ocular health and its alteration could lead to ocular disorders. Understanding the impact of intraoperative management on the ocular surface microbiome could provide insight into the potential causes of these disorders and inform future clinical practices to minimize such changes. The aim of this study is to investigate the impact of intraoperative management on the microbiota of the ocular surface before intravitreal injection and suggest the potential effects on the host's function. This research is important for understanding the importance of maintaining the health of the ocular surface microbiome and the potential consequences of disrupting it.

Materials and Methods

Data Acquisition

Ocular surface samples were collected from 41 patients (41 eyes) before intravitreal injections. The patients were divided into two groups: Group A (n = 19) received no perioperative or intravitreal management, while Group B (n = 22) received one or more of these treatments. All patients received antibiotic eye drops before the procedure. DNA was extracted from the ocular surface samples using a commercial kit (Qiagen DNeasy Blood and Tissue Kit). 16S rRNA Gene Sequencing, paired-end reads and sequences were generated using the Illumina HiSeq 2500. The marker gene data were uploaded to the National Center for Biotechnology Information's Sequence Read Archive (NCBI SRA) with access number PRJNA721101 [14] the samples were collected on the ocular surface. Operating taxonomic units (OTUs. (https://www.ebi.ac.uk/ena/browser/view/PRJNA721101)

Data Preprocessing

In this study, 41 participants were analyzed, resulting in a total of 6,301,278 raw sequence reads. These reads contained 1,666 different Operational Taxonomic Units (OTUs) with an average of 131,277 reads per OTU. After cleaning the sequences, the quality of the data was evaluated and the truncation parameter was determined. This parameter is used to determine the point where the quality of the sequence starts to decline [15]. The microbial marker-gene data was analyzed using Quantitative Insights into Microbial Ecology version 2 (Qiime2) software. To further improve the data, Deblur software was used to denoise the sequences and identify true ecological differences between taxa. The denoising process also removed any sequences that were incorrectly combined, known as chimeras, to ensure accurate results. The statistical analysis was performed using R software. The diversity and composition of

the ocular surface microbiota were compared between Group A and Group B. The differences in the diversity and composition of the ocular surface microbiota between the two groups were analyzed using the Kruskal-Wallis test and the Wilcoxon rank-sum test. P values less than 0.05 were considered statistically significant.

Taxonomic analysis

The method used in this study involved training a naive Bayes classifier using the scikit-learn library in Python. The classifier was specifically trained to identify the V3-V4 hypervariable regions of the sequences and was pre-clustered at 99 % sequence identity [16] especially deep learning, are widely applied to DTIs prediction. In this study, the main goal is to provide a comprehensive overview of deep learning-based DTIs prediction approaches. Here, we investigate the existing approaches from multiple perspectives. We explore these approaches to find out which deep network architectures are utilized to extract features from drug compound and protein sequences. Also, the advantages and limitations of each architecture are analyzed and compared. Moreover, we explore the process of how to combine descriptors for drug and protein features. Likewise, a list of datasets that are commonly used in DTIs prediction is investigated. Finally, current challenges are discussed and a short future outlook of deep learning in DTI prediction is given.","author":[{"dropping-particle":"","family":"Abbasi","given":"Karim","non-dropping-particle":"","parse-names":false,"suffix":""},{"dropping-particle":"","family":"Razzaghi","given":"Parvin","non-dropping-particle":"","parse-names":false,"suffix":""},{"dropping-particle":"","family":"Poso-","given":"Antti","non-dropping-particle":"","parse-names":false,"suffix":""},{"dropping-particle":"","family":"Ghanbari-Ara","given":"Saber","non-dropping-particle":"","parse-names":false,"suffix":""},{"dropping-particle":"","family":"Masoudi-Nejad","given":"Ali","non-dropping-particle":"","parse-names":false,"suffix":""}],"container-title":"Current medicinal chemistry", "id":"ITEM-1","issue":"11","issued":{"date-parts":[["2021"]]},"language":"eng","page":"2100-2113","publisher-place":"United Arab Emirates","title":"Deep Learning in Drug Target Interaction Prediction: Current and Future Perspectives.","type":"article-journal","volume":"28"},"uris":["http://www.mendeley.com/documents/?uuid=2cb89a42-aec2-46cb-94ba-9e79d7de2302"]}],"mendeley":{"formattedCitation":"(16. This approach allowed for the efficient and accurate analysis of the microbial marker-gene data obtained from the study. The classifier was trained using a pre-trained naive Bayes algorithm, which was trained against the GreenGenes database (13_8 revision) [17]. The diversity of phylogenetic trees and nonphylogenetic trees can also be measured as well as alpha- and beta-diversity. Alpha diversity is a measure of the diversity within a single sample or community and beta diversity is a measure of the diversity between different samples or communities [18]. In order to calculate alpha diversity, the Shannon and Brey-Curtis indexes were used. The Shannon index is a measure of the diversity of a community and takes into account both the number of different species present and their relative abundance [19]. The Brey-Curtis index is a dissimilarity measure that compares the relative abundance of different species in two communities. The Jaccard index is a measure of similarity between samples based on the presence or absence of different species.

Table 1: Groups of patients and their demographic characteristics

Parameter	Group A	Group B
Number of samples	19	22
Age (year)	61.53 ± 14	63.3 ± 9.19
Gender composition (Male-Female)	8 + 11	14 + 8
Disease (sample number)		
diabetic retinopathy	12	7
age-related degeneration	4	7
center retinal vein occlusion	2	0
polypoidal choroidal vasculopathy	1	3
branch retinal vein occlusion	0	2
neovascular glaucoma	0	3

In this study, various measures were used to assess the diversity of the microbiome samples. Alpha diversity measures, such as phylogenetic diversity, Shannon diversity, evenness, and observed OTUs were calculated. Beta diversity measures, such as weighted/unweighted UniFrac, Bray-Curtis, and Jaccard were also determined. QIIME2 software was used to visualize the beta diversity measures. The analysis focused on the mean abundance and prevalence of each taxon. Random sampling was utilized to calculate the mean prevalence and abundance of each OTU, as well as the upper and lower confidence intervals. The Kruskal-Wallis test was used to evaluate significant differences between groups based on alpha-diversity values, while a PERMANOVA was employed to assess differences in beta-diversity between groups. Additionally, alpha rarefaction analysis was utilized to determine if sufficient depth had been sequenced to capture most taxa in the environment [20]. This was achieved by subsampling a sequence of depths at random and plotting the depth of sequencing as a function of alpha diversity measures computed from these subsamples [11].

Results

The results of the study showed that the sequence diversity among the 41 patients was substantial, with a range of 14610 to 47201 sequences per sample and a mean of 31979.57 sequences per sample. These sequences were clustered into 2541 different operational taxonomic units (OTUs). The demographics of the patients, including age, gender, and disease type, are presented in Table 1. Importantly, there were no significant differences between the groups in terms of sequence diversity ($P > 0.05$). This suggests that the diversity of the gut microbiome is not strongly influenced by factors such as age or gender.

The schematic diagram depicts the pipeline used to analyze the data in Figure 1.

Rarefaction analysis

The analysis of rarefaction shows that as the number of sequence samples increases, fewer new OTUs are detected. At the 14500OTUs

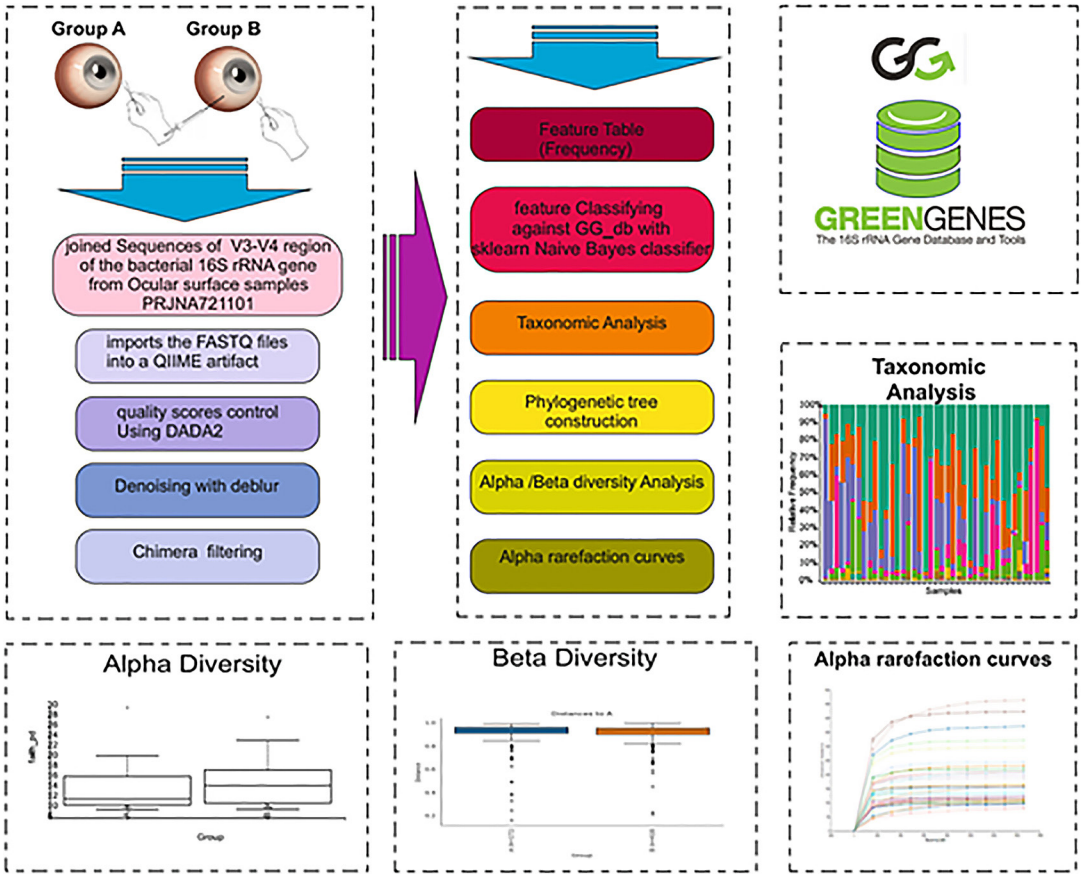

Figure 1: Pipeline that was used in this study

level (the depth of analysis used in this study), at this point, the curves for most samples have plateaued (Figure 2), indicating that additional sequencing would be unlikely to reveal novel OTUs.

Microbial Diversity Analysis

The result from our analysis shows that there is a significant difference in the alpha diversity of microbial communities between the samples based on the clustering results of all OTUs. Evaluation indices such as Simpson, Chao1, ACE, Fisher, and Shannon were used to analyze the alpha diversity, and the results indicate that there is a statistically significant difference between the samples ($P < 0.05$; Figure 3-a). Additionally, the study used a non-phylogenetic beta diversity metric, the Bray-Curtis analysis, to compare the groups at the genus level. The results of this analysis show a significant difference in microbiota composition between Groups A and B ($P = 0.014$; Figure 3-b). This suggests that the microbial community compositions of the two groups are distinct from one another.

Microbiota Composition on the Ocular Surface:

The results showed that there was a comparison of microbiota abundance among the groups based on phyla and genera. This comparison was presented in Figure 4a and 4b, which showed the ten most important microbiota in each group. The beta diversity analyses were used to measure differences between groups, and the results showed that at the phylum level, Proteobacteria and Actinobacteria had greater relative abundance compared to

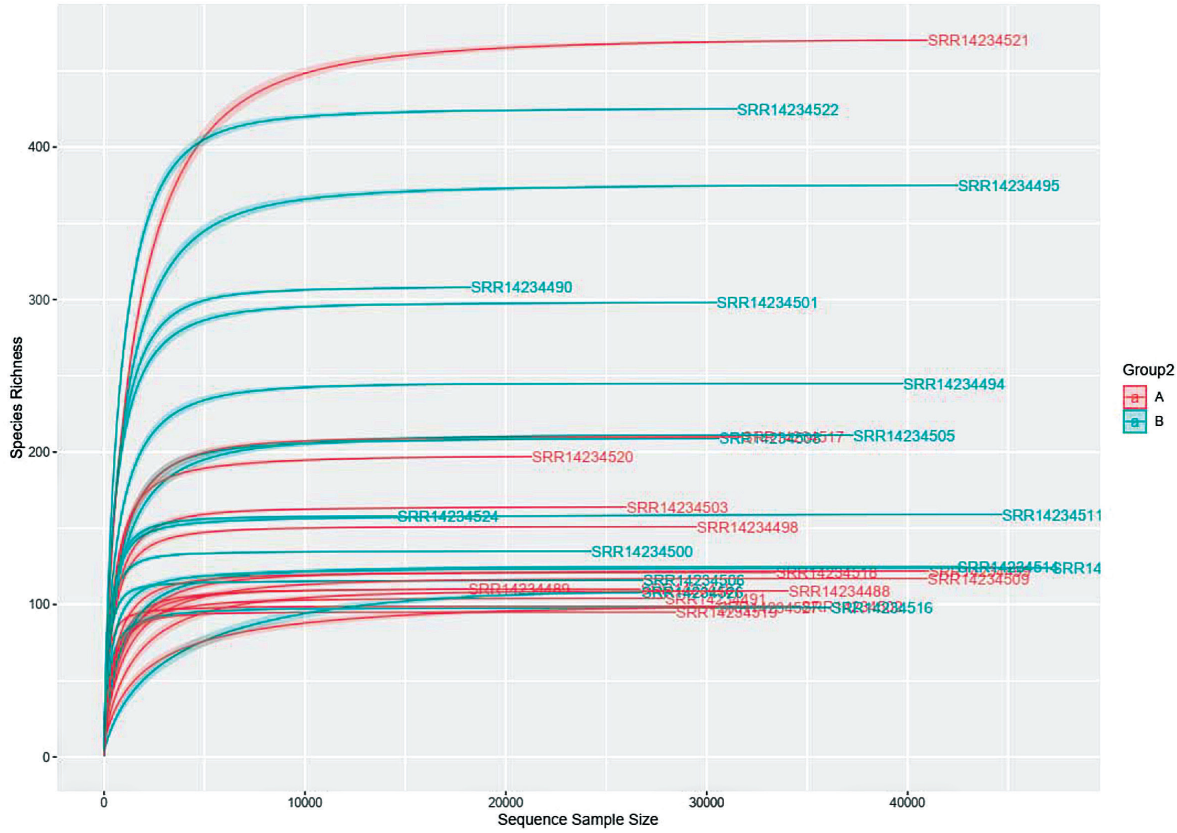

Figure 2: Alpha rarefaction curve for each sample, Initial rapid increase in alpha diversity with increasing sequence depth. Leveling off of alpha diversity at a certain sequence depth, indicating that most taxa have been captured in the sample

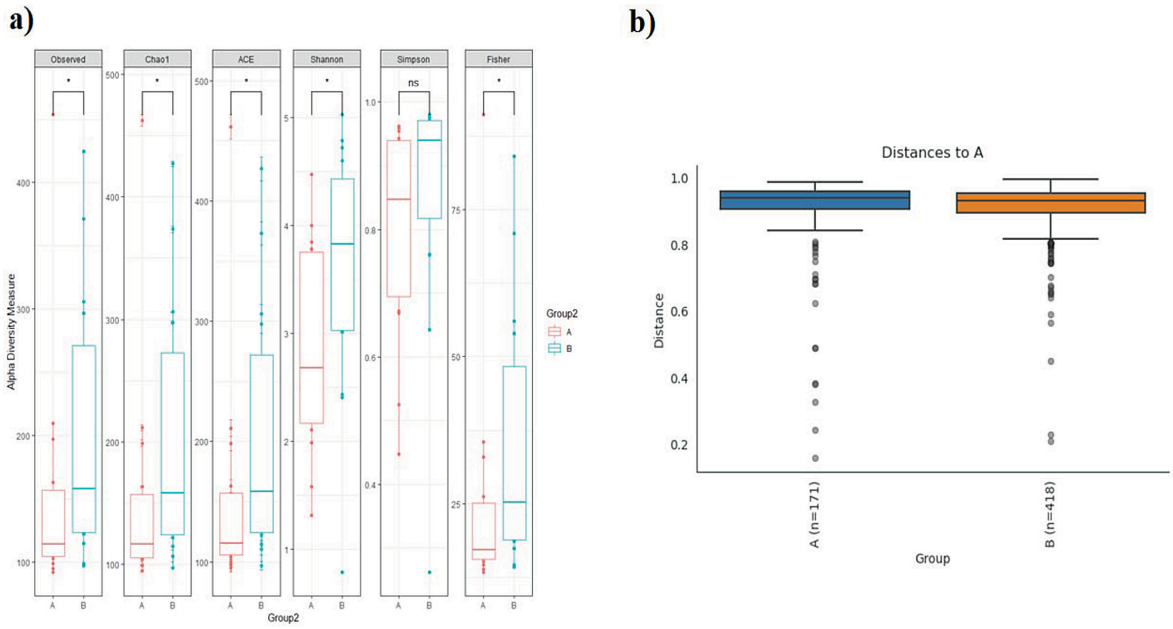

Figure 3: a) Alpha diversity, b) Plots of group significance: To assess differences between the groups, beta diversity analysis was used

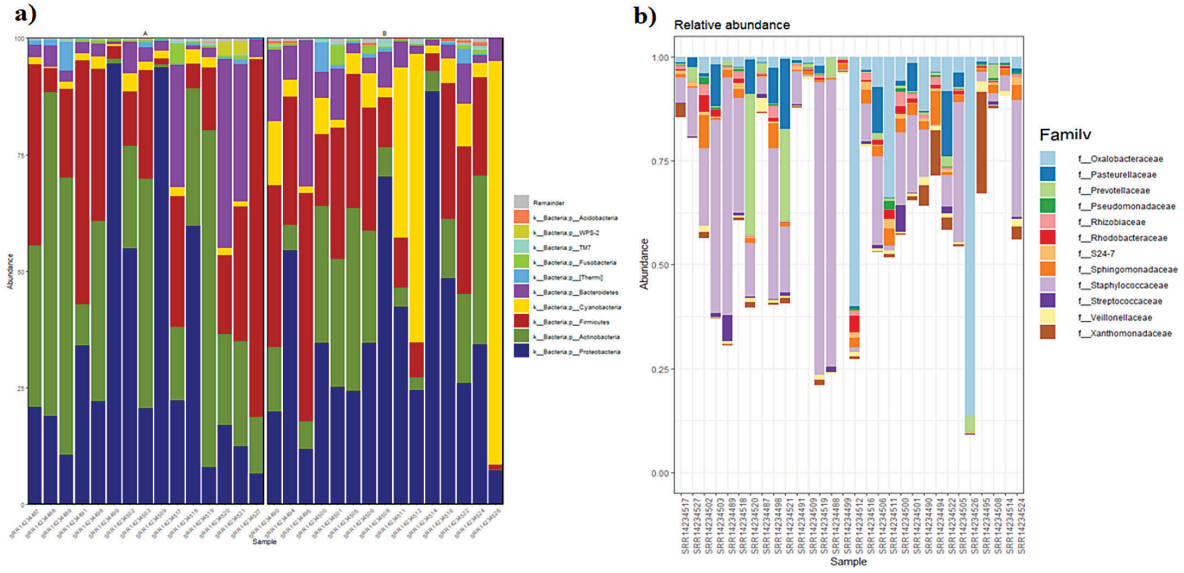

Figure 4: a) Comparing phylum-level microbiota abundances, b) Comparing family-level microbiota abundances

other phyla. The group B showed an increase in Actinobacteria and a gradual decrease in Proteobacteria. Cyanobacteria also sharply increased, while Firmicutes and Bacteroidetes were little affected. These results suggest that perioperative management has clearly altered the microbiota on the ocular surface, as demonstrated by the relative abundances at the family level shown in Figure 4b.

Analysis of Microbiota Differences:

A significant difference was observed between Group A and Group B in the number of bacteria like Facklamia, Proteus, Aquabacterium, Pelomonas, and Sphingopyxis. Moraxella prevalence differed significantly between Group A and Group B. Ocular microbiota had been altered by perioperative management (Figure 5).

Additionally, an analysis was conducted for identifying the relationship between abundance and prevalence, which is shown in Figure 6. Taxa that are highly prevalent in a group of samples can be identified by examining their average relative abundance. High prevalence taxa can be identified at varying values of mean relative abundance with low variation in lower and upper confidence intervals. It is most likely that these taxa are the core taxa found in the Actinobacterial, Bactericides, Cyanobacteria, Firmicutes, and Protozoa groups of samples.

Phylogenetic tree

This method of making evolutionary trees, also known as phylogenies, illustrates the evolutionary branches from which different species, creatures, or genes descend. In evolutionary terms, it is situated on the same branch as species, creatures, or genes. The tree includes a dot next to every tip (OTU). By using the top 50 taxa in figure 6, we can visualize the top 50 taxa better.

Discussion

Most ophthalmologists agree that antibiotic eye drops can reduce the risk of infectious endophthalmitis. Studies suggest that antibiotic eye drops applied to the ocular surface are not a preferred method for preventing intraocular

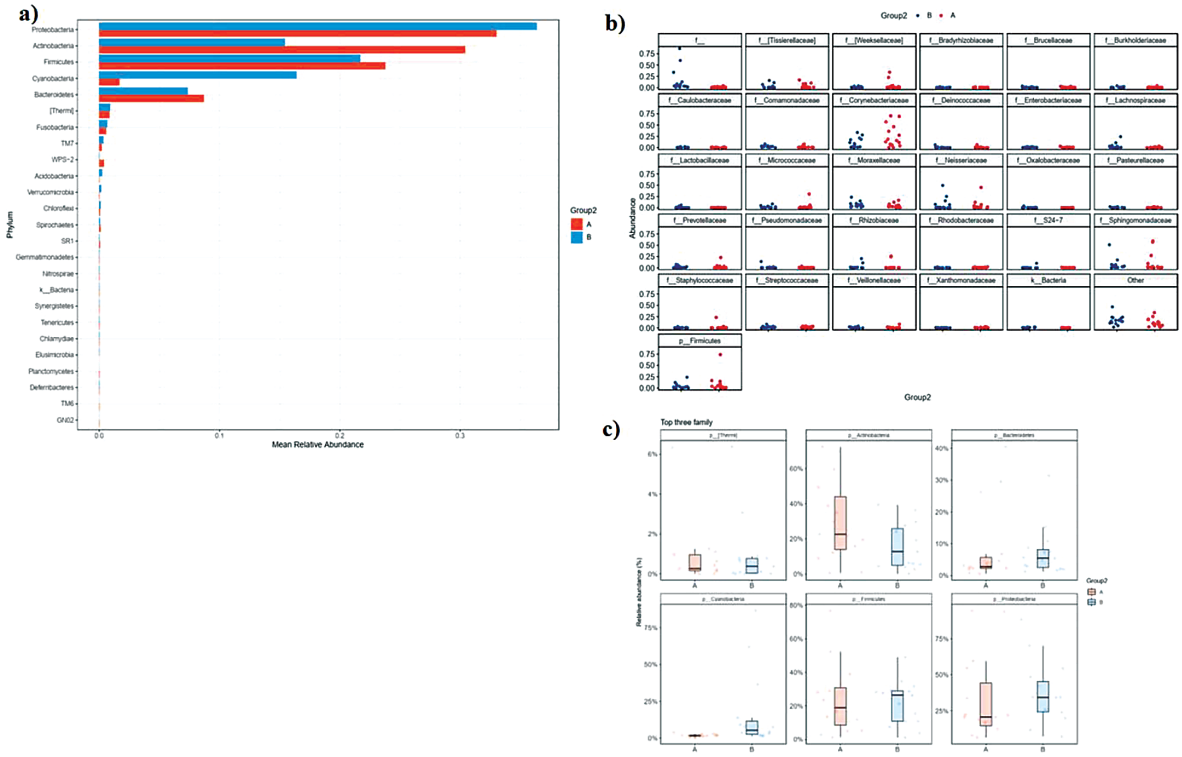

Figure 5: a) Mean relative abundances of different microbiota between two groups at the phylum level, b) top three families that decreased and increased in group B, c) top three family's relative abundances in two groups

infections [21]. One possibility is that the antibiotics used in prevention are not effective against the specific microorganisms present in the preventing intraocular. A second is possible that the antibiotics are not reaching the target area effectively due to inadvertent administration of the antibiotics or microbial resistance mechanisms. Additionally, antibiotics can disrupt the balance of the microbial community, leading to the overgrowth of pathogenic species and the suppression of beneficial species. This can increase the risk of ocular surface infections and inflammation, even if the incidence of endophthalmitis is not affected, it is essential to consider the potential negative impact of antibiotic use on the ocular surface microbiome. [22 14, 23] bacterial infections have again become a threat because of the rapid emergence of resistant bacteria-a crisis attributed to abuse of these medications and a lack of new drug development.","author":[{"dropping-particle":"","family":"Ventola","given":"C Lee","non-dropping-particle":"","parse-names":false,"suffix":""}],"container-title":"P & T : a peer-reviewed journal for formulary management","id":"ITEM-1","issue":"4","issued":{"date-parts":[["2015","4"]]},"language":"eng","page":"277-283","publisher-place":"United States","title":"The antibiotic resistance crisis: part 1: causes and threats.","type":"article-journal","volume":"40"},"uris":["http://www.mendeley.com/documents/?uuid=71ff24bd-ad10-44e1-bd5a-ea78a1f75aa2"]},{"id":"ITEM-2","itemData":{"DOI":"10.18240/ijo.2022.02.09","ISSN":"2222-3959 (Print. Using

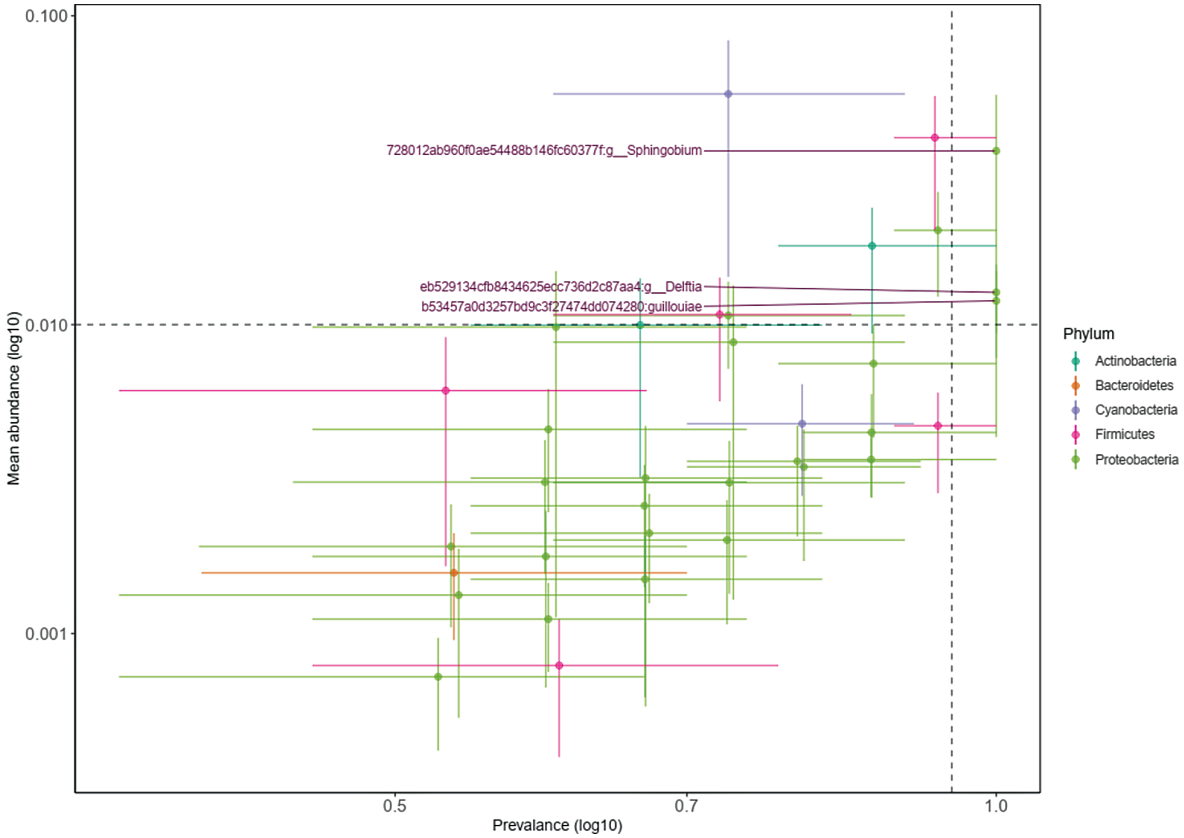

Figure 6: Identification of a core Microbiome

antibiotic eye drops before intravitreal injection changed the composition of ocular surface microbes. There is an increase in gram-negative bacteria in infectious endophthalmitis [24, 25]. As nonpathogenic and symbiotic microbes become more abundant, pathogenic bacteria may grow, causing endophthalmitis and bacterial keratitis, which can be devastating. As nonpathogenic and symbiotic microbes become low abundant, pathogenic bacteria may grow, causing endophthalmitis and bacterial keratitis, which can be devastating. Furthermore, Our results show there were no significant differences in the incidence of infectious endophthalmitis between patients who received antibioprophylaxis and those who did not, and These findings suggest that perioperative management, specifically the use of antibioprophylaxis, impacts the ocular surface microbiota. Clinical studies have shown that most pathogens are common in nearly 75 % of cases, and gram-negative bacteria caused intraocular infections [26]. Also, gram-positive bacteria commonly cause infections on the ocular surface [27–29].

An analysis of the structure and diversity of the ocular microbiota was conducted in the present study. The microbiota on the surface of the eye was detected following perioperative management and intravitreal injections. Microbiome diversity is measured by alpha diversity. Beta diversity between groups is a measure of macrobiotic diversity.

Within each group, alpha diversity was different, and between Groups A and B, beta diversity was different. Based on these results, perioperative management significantly altered ocular microbiota in each group. A

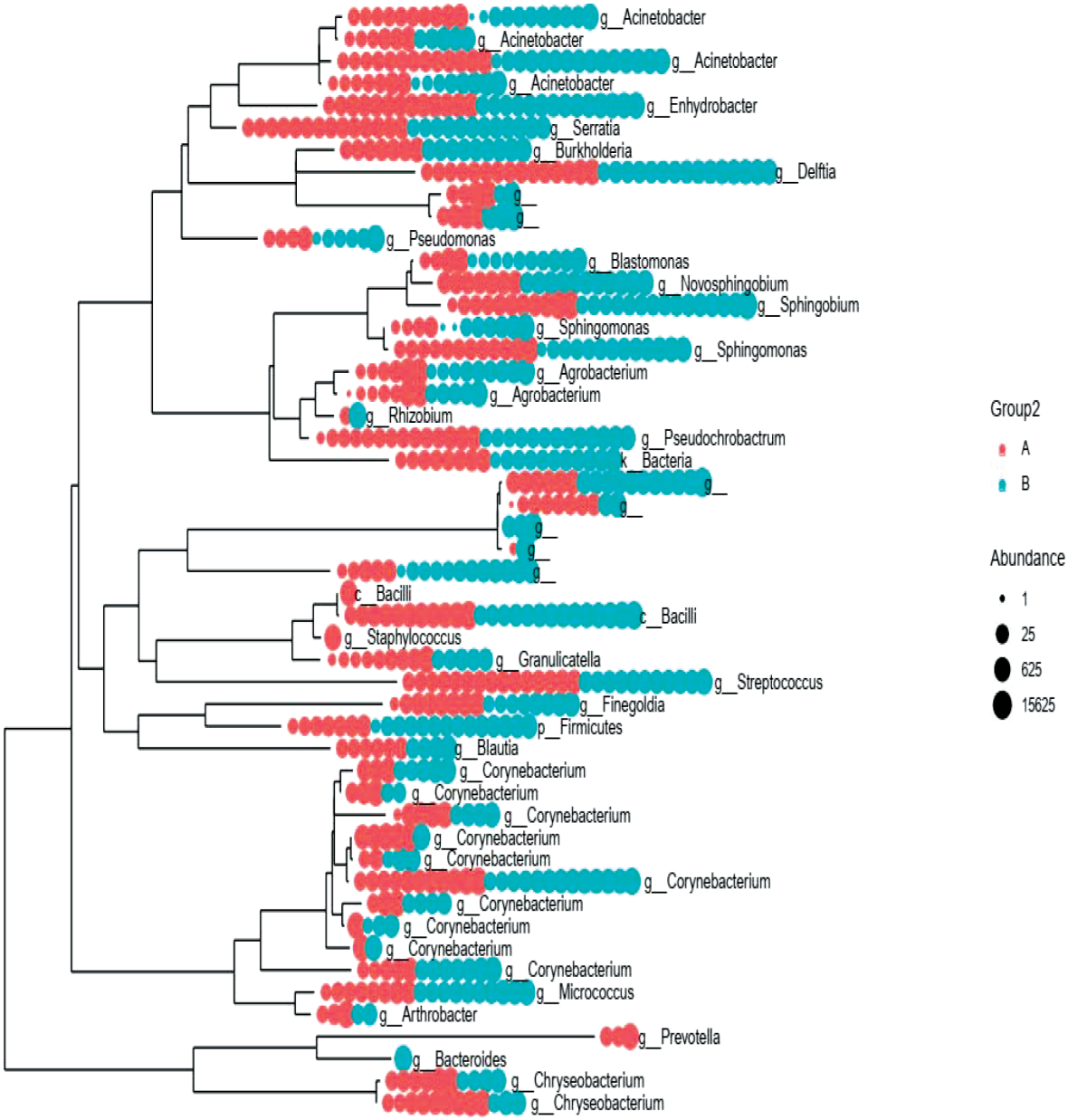

Figure 6: Top 50 taxa for better visualization of tree

significant change in the composition of the ocular surface microbiota was also observed as a result of intravitreal injections. During perioperative procedures, using local antibiotic eye drops may disrupt the original balance of the ocular surface microbiota, thereby adversely affecting the ocular surface's homeostasis.

In this study, both groups showed the highest abundance of Bacteroidetes, Cyanobacteria, Firmicutes, Actinobacteria, and Proteobacteria at the phylum level. Cyanobacteria and Actinobacteria showed the greatest variation among these phyla. A systematic review [30] that used 16S rRNA sequencing to identify the most prevalent bacteria based on their phyla found that Corynebacterium was present in the eye microbiome in all publications, with a median percentage of 10 %. Other commonly found bacteria included 66 % of Corynebacterium,

19 % of Pseudomonas, 66 % of Staphylococcus, 77 % of Propionibacterium, and 3 % of Streptococcus. Although Bacillus was not included in the core microbiome, it was present in 8 % of cases when present. The results suggest that the most abundant phyla are Proteobacteria (Pseudomonas), Actinobacteria (Corynebacterium and Propionibacterium), Firmicutes (Staphylococcus and Streptococcus), and Bacteroidetes. These findings indicate that perioperative management that changes the microbiota on the ocular surface.

CONCLUSION:

Ocular surface microbiota becomes imbalanced due to perioperative management, which alters the relative composition and abundance of microorganisms. Furthermore, perioperative management alters the composition of microbiota on the ocular surface by disrupting the balance. Physicians should consider the implications of perioperative management more carefully before administering intravitreal injections. Gram-negative bacteria were also increased in the eye following intravitreal injections prior to perioperative management. Finally, clinical doctors need to pay more attention to the effects of intravitreal injections during the perioperative period.

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Authors ORCIDs

Hamidreza Taherkhani:
https://orcid.org/0000-0002-5853-7260
Azadeh Kavianfar:
https://orcid.org/0000-0003-0880-0291

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Footnotes and Financial Disclosures

Conflict of interest:

The authors have no conflict of interest with the subject matter of the present manuscript.