NSU
Florida

College of Dental Medicine
NOVA SOUTHEASTERN UNIVERSITY
Research Day
June 6-7, 2023

Nova Southeastern University College of Dental Medicine

2023 Research Day

Dear Students, Colleagues, and Friends:

Welcome to the inaugural 2023 Nova Southeastern University College of Dental Medicine's Research Day, an annual event that showcases the outstanding research conducted by our graduate/post graduate students and faculty. In the following pages we are pleased to share with you the abstracts from these research projects that progress the field of dental science.

With support from Nova Southeastern University's Mission, Vision and Core Values, we at the College of Dental Medicine aspire to be a leader at the University and globally by our commitment to high quality dental research.

We take this opportunity to thank our very talented and dedicated faculty who each year mentor and guide our students in their research projects. We are grateful to Nova Southeastern University and the Health Professions Division for the opportunity to conduct research by students and faculty and making these projects possible through several grant programs.

Some of these projects also represented the College of Dental Medicine at National and International meetings.

Congratulations to the students and faculty presenting in the 2023 CDM Research Day.

Dr. Steven I. Kaltman
Dean and Professor

Dr. Sibel Antonson
Professor, Assistant Dean for Research and Development

Dr. William B. Parker
Associate Professor & Associate Dean for Postgraduate Education
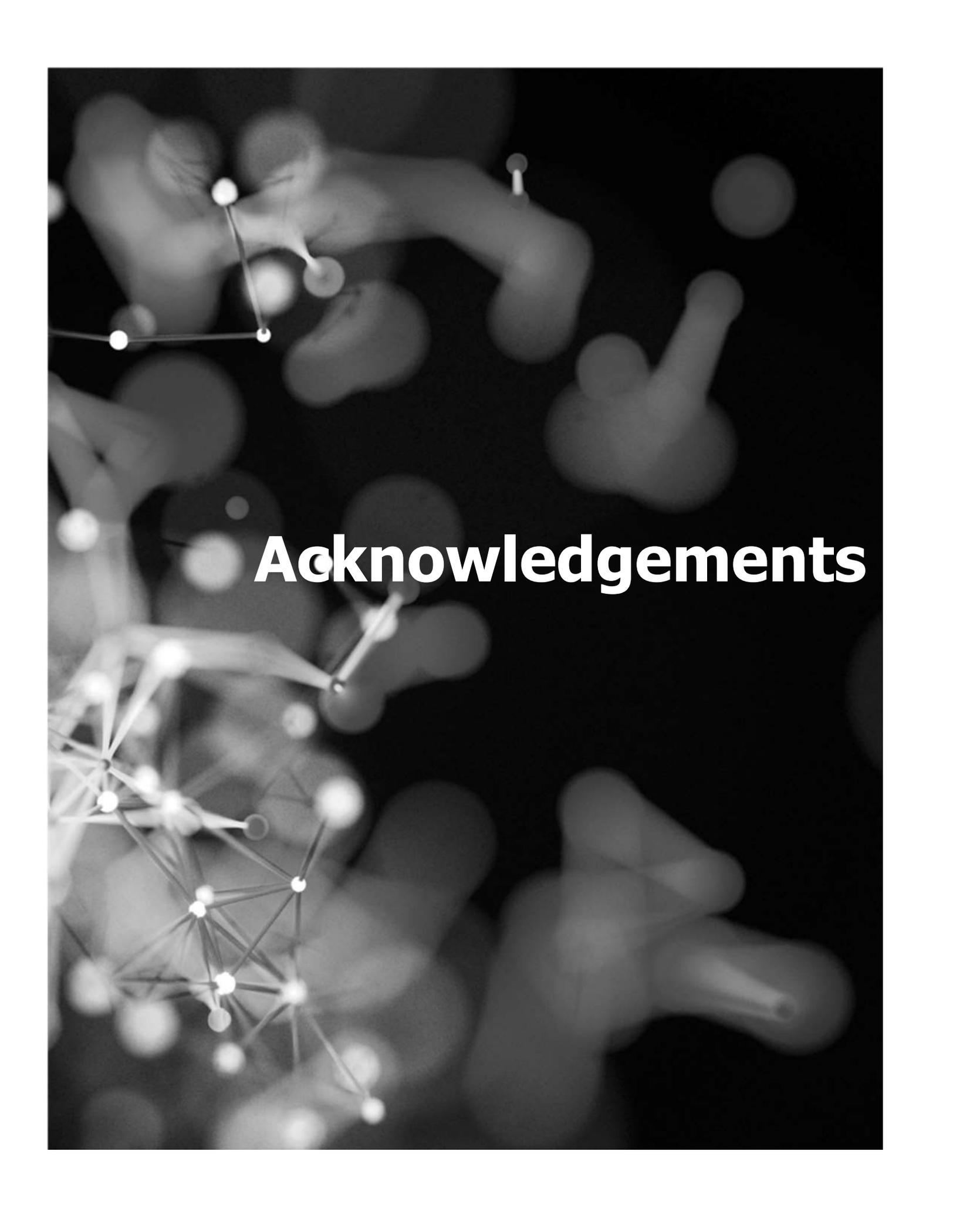
Acknowledgements

***A special thank you and show of gratitude to the following doctors
that gave their time, expertise and their knowledge to the
reviewing process that contributed immensely
to the CDM 2023 Research Day.***

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Bittner, Nurit

Bontempo, Alexander

Feit, Daniel

Huang, Shengyuan

Leon, Elizabeth

Mesko, Mauro

Molina, Cesar

Pastore, Maria Rita

Siegel, Michael

Suzuki, Maiko

Vardar, Saynur

**Keynote Speaker
2023 June 6**

FROM POSTER TO PUBLICATION – PATHWAY GUIDELINES

This short presentation will review important steps to be taken to enhance/organize the information you have already presented as a poster at the Research Day event, and submit it to a peer-reviewed, index dental research Journal.

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Dr. Rueggeberg received his dental degree from Emory University, after which he completed a general dental residency program in Wilmington, DE. He then associated with a dentist and later established a private, general dental practice in Seaford, DE. He received a master's degree in Biomaterials from The University of Michigan in 1987 and began his academic career at the then Medical College of Georgia School of Dentistry. He is currently a retired/rehired Emeritus Professor in the Department of Restorative Sciences at the Dental College of Georgia at Augusta University. His research interests focus on photo-polymerization of dental resin systems – restorative materials and ancillary dental equipment. He has authored or co-authored 226 peer-reviewed articles and 263 abstracts and has lectured nationally and internationally in the field of dental photo-activated materials and many other topics at 174 separate events and works with industry in development and testing of new products and equipment. He has mentored numerous students (pre-dental, dental, resident, and graduate) as well as dental faculty and international visiting students/faculty and is founder and co-administrator of The Department of Restorative Sciences' program called FOSTER: Faculty Opportunity Strategies Towards Effective Research. He is an inventor or co-inventor of 8 full US patents and 4 provisional patents covering a variety of areas used in clinical dental materials. He and his wife, Sallie, recently celebrated their 28th wedding anniversary, and have three children (2 boys and one girl) and 5 grandchildren (4 girls, one boy). However, the LOVES of their lives and consumers of the majority of their income and time are their six 4-legged "children" (5 chihuahuas and one confused American Boxer/mix).

Presentations
2023 June 6

DIGITAL ASSESSMENT OF PROTECTIVE EFFECT OF FLUORIDE VARNISHES AGAINST ENAMEL WEAR AFTER DEMINERALIZATION/TOOTHBRUSH ABRASION

H. Albagami, BDS, MS, PhD; E. Kilinc DDS, PhD, MPH; C. E. Garcia-Godoy, DDS, MPH; P. Hardigan, PhD; S. Antonson, DDS, PhD, MBA

Objectives: To evaluate the protective effects of various fluoride varnishes on enamel subjected to pH-cycling and toothbrush abrasion via digital measurement of enamel surface loss and wear.

Methods: 140 specimens were prepared from extracted human teeth. Specimens were divided into six varnish groups (Fluor Protector S, Vanish, NUPRO White, ProFluorid, Duraphat, PreviDent), and a Control group–no varnish (n=20/group). Varnishes were applied according to manufacturers' instructions. Specimens were subjected to an 8-day pH cycling, then exposed to 500 and 1,500 cycles of toothbrush abrasion to simulate one and three-months of brushing respectively. 3D digital scans were obtained using an intra-oral scanner (CEREC OmniCam) at baseline, after 500 and 1,500 cycles of brushing. Wear measurement and analysis were conducted using exocad software. To compare differences for the enamel-wear outcome measure, a two-way ANOVA model with interaction was created.

Results: After 500-cycle abrasion, the Control group(51.28±10.96μm) showed significantly higher enamel wear than varnish groups ($p<0.001$), except for ProFluorid(44.44±13.63μm)($p=0.341$). Fluor Protector S(27.16±5.08μm) displayed the most resistance to brushing abrasion. After 1,500 cycle abrasion, Control group showed significantly more enamel wear than varnish groups (64.72± 13.49μm)($p<0.001$). Fluor Protector S(33.42±5.13μm) demonstrated the most resistance to brushing abrasion, showing significantly less enamel wear than Prevident (45.00± 8.96μm)($p=0.007$), Vanish (44.74± 10.45μm)($p=0.009$), ProFluorid (51.79± 18.83μm)($p<0.001$), and Control ($p<0.001$) groups. However, no significant differences were detected between Fluor Protector S and Duraphat (33.28± 5.53μm)($p=1.0$) or NUPRO White (40.04±11.37μm)($p=0.386$).

Conclusions: Fluoride varnishes demonstrated a significant protective effect on the enamel surfaces. However, not all fluoride varnishes performed equally, despite most of them sharing the same active ingredient. Unprotected enamel is prone to wear as a result of demineralization and toothbrushing, and the application of fluoride varnish appears to reduce the amount of demineralization, although results vary by varnish.

CURRENT IRRIGATION PROTOCOL USED IN ENDODONTIC OFFICES: A 2022 SURVEY

Mahzad Koochaki, DMD; Jason Zeim, DMD

Introduction: The goal of this study was to evaluate the most common irrigation methods and solutions currently being used by endodontist members of American Association of Endodontists (AAE).

Methods: A survey consisting of 16 questions was emailed to 3,398 active endodontist members of AAE. The survey included questions regarding the solutions, percentages and methods used in irrigation of root canal systems. The survey had a response time interval of two months.

Results: The total number of respondents included 523 participants. The average age of respondents was 47.20 with an average of 15.85 years in practice. 79.73% of respondents work in private practice and 40% of the respondents were board-certified. 89%,85% of respondents use NaOCl, EDTA respectively. Other most commonly used solutions included Chlorhexidine (64%), Saline (61%) and Chloroform solution (60%). 6% NaOCl and 17% EDTA was the most commonly used concentration. 23.3% of respondents did not know the percentage of NaOCl used for their treatment. The order of solution used for irrigation included NaOCl followed by EDTA. 84.7% of respondents remove smear layer, and the most commonly used solution was EDTA. The average irrigation time for root canal therapy by most of the respondents was 30 minutes. The most common irrigation delivery system included needle delivery (56.4%). 30-gauge side-vented needle was the most common needle used for irrigation with 10-15ml volume of irrigation delivered by most respondents. 16.3% of respondents use commercial irrigation system, and 8% use laser for root canal disinfection.

Using a Chi-Squared analysis with a significance level of <0.05 (p < 0.000), there was a significant difference between the proportions of used irrigation solvents. Sodium hypochlorite (NaOCl) and ethylenediaminetetraacetic acid (EDTA) were the most commonly used solutions among endodontist members of AAE.

Using a Chi-Squared analysis with a significance level of <0.05 (p < 0.000), there was a significant difference in the preferred irrigation delivery methods. The traditional irrigation needle delivery system was the most commonly used system among endodontist members of AAE.

Conclusions: NaOCl was the most common irrigation solution used by endodontist members of AAE, followed by EDTA. The most common irrigation delivery system was needle with average irrigation time of 30 minutes and 10-15ml total volume delivery.

The most commonly used irrigant solutions used by endodontist member of AAE

Most common used irrigants
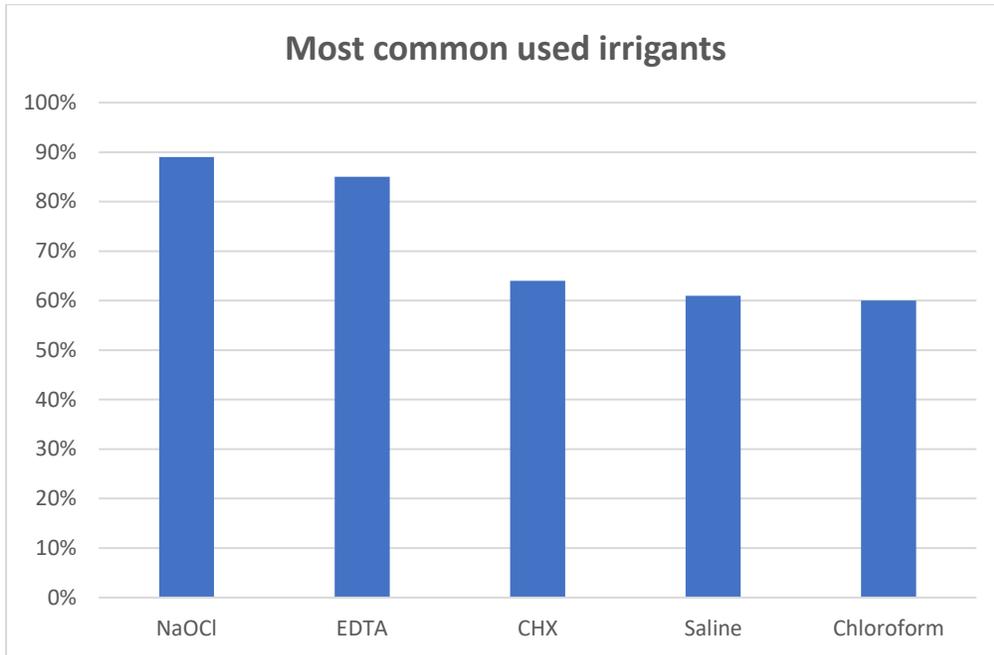
The percentage of endodontist member of AAE using various concentrations of NaOCl
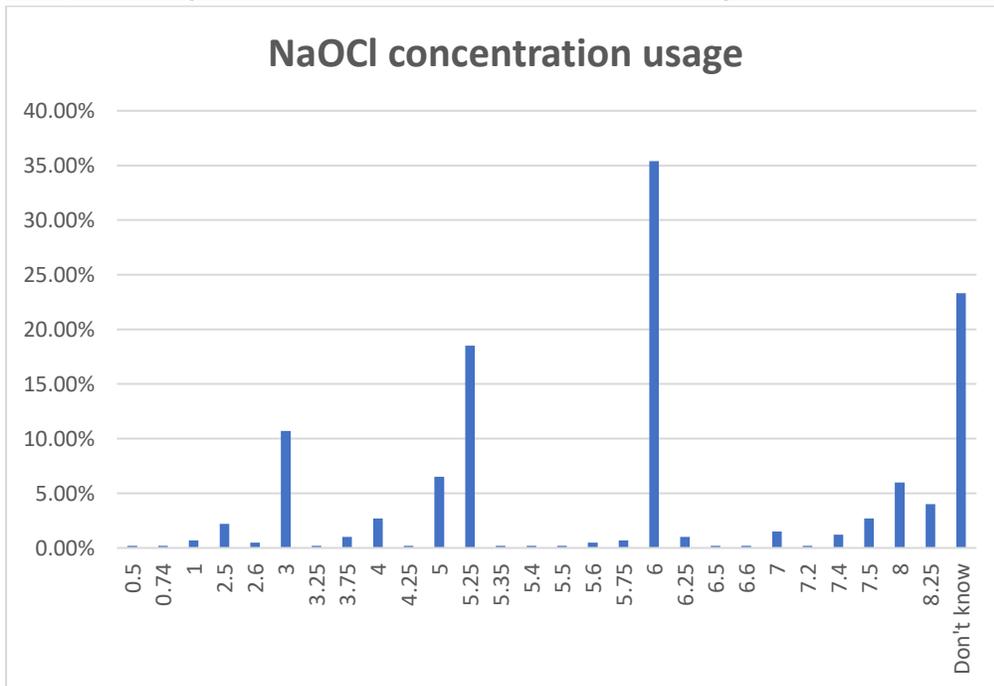
NaOCl concentration usage

WELL-DIFFERENTIATED SQUAMOUS CELL CARCINOMA ARISING FROM A PRE-EXISTING ODONTOGENIC KERATOCYST

Matthew Kelley, DMD; Jason Portnof DDS, MD.

Introduction: The term "primary intraosseous squamous cell carcinoma" was introduced in 2005 by the World Health Organization with three subcategories. Squamous cell carcinoma arising from the lining of an odontogenic cyst is one important rare subcategory of such lesions with an incidence of 0.01 to 0.02%. Furthermore, the appearance of such malignancy in an odontogenic tumor such as odontogenic keratocyst is considered extremely rare.

Case: In this case report, we review a case of a 74-year-old Caucasian male who reported a several year history of left mandibular swelling and expansile growth. The patient's mandibular swelling progressively increased until the patient began experiencing hypoesthesia of the left inferior alveolar nerve distribution. Upon initial clinical and radiographic examination, the patient was noted to have a large radiolucent cystic lesion in the left posterior mandible. The initial biopsy of the cyst was obtained on 11/25/2014 and returned as an orthokeratinized odontogenic cyst with focal chronic inflammation. A secondary confirmation biopsy was obtained on 1/25/2015 which returned as an odontogenic cyst with epithelial dysplasia with luminal ortho and parakeratin formation. The patient was lost to follow up until 2023 until he presented to our clinic. At that time, the patient underwent an additional biopsy of the lesion which returned as an odonogenic keratocyst with dysplasia. Due to the size and aggressive nature of the lesion, the patient elected to proceed with segmental resection of the mandible as definitive treatment for the cyst. The final pathology of the cystic lesion was consistent with well differentiated squamous cell carcinoma arising from a preexisting odontogenic keratocystic tumor.

Conclusion: Primary intraosseous squamous cell carcinoma arising from a keratocystic odontogenic tumor is considered to be very rare. *Bodner et al.* highlighted the presence of only 16 cases of PIOSCC arising from KCOT from 1938 to 2010. Even though SCC arising from KCOT is a rare phenomenon, the present case is comparable with most of the aspects cited in the literature. Data on the prognosis of PIOSCC arising from KCOT is scarce as there are only a few reported cases. However, in the reported cases, the 2-year survival rate of patients has been 53%. The prognosis was reported to be poor when there was evidence of nodal metastasis of the neck. Surgical management was recommended in most reports with adjuvant radiotherapy. The case emphasizes the importance of careful investigation of swellings present in the mandible as well as the timeline associated with treatment or lack thereof. Clinicians as well as patients should be aware and detect these changes to avoid being clinically negligent and to prevent poor patient outcomes.

ANGIOMATOID FIBROUS HISTIOCYTOMA OF THE MANDIBLE:
A CASE REPORT AND REVIEW OF LITERATURE

Maria Nord, DDS; Victor Celis, DMD; Mark Mintline, DDS; Shawn McClure, DMD, MD

Objectives: To publish a review article on the current literature on angiomatoid fibrous histiocytoma (AFH) and present a case study in which it presented intraosseous in the mandible of a young, female patient.

Background: AFH is a relatively rare sarcoma that typically affects the somatic tissues of extremities. It is relatively rare in the head and neck, and when diagnosed in this region, little literature exists which discusses behavior of the tumor, treatment modalities, complications, and recurrence rates. Overall, AFH is said to have good prognosis with local recurrence being around 12% and only rarely metastasizing.

Results: A review of literature was completed using: Medline, EMbase, and Google Scholar databases with keywords: angiomatoid fibrous histiocytoma, fibrous histiocytoma, AFH, head and neck sarcoma, benign fibrous histiocytoma, malignant fibrous histiocytoma. Very few articles exist that reference Angiomatoid Fibrous Histiocytoma (AFH). Fifteen articles were included. Reported ages ranged from 6 months to 83 years. The size of the masses ranged from 0.5 cm to 10 cm. In our research, 49 were male, 41 female and one individual's sex was not specified. The lesion was treated successfully by segmental mandibular resection, and perhaps this case can aid in both the diagnosis and treatment of these lesions with bony invasion of the jaws.

Case: A 9-year-old African American female with no relevant medical history presented with a mild tenderness of the right mandibular angle that reportedly started two days prior to presentation. On examination, the patient had a firm, non-erythematous, mass localized to the right mandibular angle. On CT neck soft tissue with IV contrast, a 1.9 x 2.4 x 1.9 cm mixed cystic and solid lesion was identified at the level of the right mandibular angle. She was taken to the operating room for an attempt at an incisional biopsy was performed. A soft tissue specimen of the lesion was sent for histologic evaluation. The specimen was sent for expert consultation where it was described as a 2.8 cm, atypical epithelioid neoplasm with cystic change. In this report the mass was described as having cysts filled with blood and surrounded by hemosideran-laden macrophages. This second pathologist stated there was extensive lymphoid response at the periphery associated with fibrosis which they believed to be a rare example of AFH involving bone with a soft tissue extension.

Conclusions: Based on our study and previous studies, we can expect variable desmin, CD99, and EMA immunoreactivity in AFH histology. Desmin, epithelial membrane antigen, is positive in about 50-60% of cases. In our case, desmin was focally positive. Additionally, all other essential histologic (H&E) criteria were present in our patient's tumor as described histopathologically. Based on current literature and our study, prognosis seems to be affected by the ability to obtain negative margins during resection. We were able to achieve negative margins in our case, likely due to early presentation, diagnosis, and treatment. Our patient is now 15 months status post mandibular resection with no signs or symptoms of recurrence. She maintains regular follow up visits.

PARENT-IDENTIFIED PERCEPTION OF CHILD'S BEHAVIOR DURING THE INITIAL DENTAL VISIT AMONG CHILDREN WITH AUTISM SPECTRUM DISORDER:
A CROSS SECTIONAL STUDY

Chanin, M., Etcheverry, N., Levi-Minzi, M., Chung, J., Padilla, O., Ocanto, R.

Objectives: The purpose of this study was to verify parent perception of behavior, to determine parent ability to predict their child's dental treatment cooperation during the first dental visit, and to determine the "success" of the first dental appointment for patients with Autism Spectrum Disorder (ASD).

Methods: Pre-Treatment form data, Task Analysis Scores (TAS), and Frankl Scale ratings were extracted from patient charts available in axiUm (academic dental software). Frequencies and mean values were calculated for patient demographics and health characteristics (N=235). Multiple regression models were constructed to examine the productivity (success) level achieved during the first dental appointment (as measured by TAS and Frankl scores) by the following factors: patient demographics, parental perception of behavior, past dental visits, dental needs, and behavioral management techniques.

Results: In the model to test patient characteristics including age, gender, ethnicity, and verbal communication, Hispanic ethnicity significantly predicted TAS score, $F(4, 191) = 2.45$, $P=.05$ [95% CI -17.18, -3.53], and age significantly predicted Frankl score $F(4, 194) = 5.17$, $P=<.001$ [95% CI 0.04, 0.12]. There was a significant association between caregiver perception of behavior and Frankl scores, $F(2, 202) = 7.68$, $P=<.001$ [95% CI -0.11, -0.03].

Conclusion: The results indicate that ethnicity and age play a role in a successful outcome during the first appointment. Additionally, parent perception of their child's behavior significantly predicted Frankl score, suggesting that coordinating with parents during the dental appointment can be beneficial in treatment planning for productive dental visits among children with ASD.

EVALUATING OUTCOME METRICS AND DIRECTIONS IN ORTHODONTIC CASE REPORTS AS PUBLISHED IN AJO-DO FROM 1966 TO 2021

Dr. Emilio R. Hernandez, DMD.

Objective: The objective of this study is to determine if there is linearity in the trend of reported records and reported positive outcomes in the case reports published in AJO-DO. The findings from this study can inform orthodontics professionals about what aspects of orthodontic case reports need to be improved for completeness.

Background: Case reports are viewed as the first line of evidence for patient care and in the field of orthodontics, these are easily accessible in the American Journal of Orthodontics and Dentofacial Orthopedics (AJO-DO). The AJO-DO sets out guidelines for case reports which includes: the uniqueness of the case, the completeness of the records, the quality of the records, the quality of the treatment, and the quality of the manuscript. According to the American Board of Orthodontics (ABO) and previous research completed by Björk, Tweed, Little, Ricketts and others, treatment outcomes can be measured with various metrics. These metrics include: the mandibular plane angle, arch form, intercanine distance, intermolar distance, lower incisor position, ideal overjet and overbite, class I canine relationship, class I molar relationship, and lip position. Using the previously mentioned metrics, this study reviewed published case reports to evaluate if there is a trend of outcome directions between finished cases in AJO-DO and the generally accepted orthodontic knowledge of Tweed, Ricketts, Little, and Björk. The approach of this study is innovative itself as it is the first to evaluate the long-term trends of outcome directions in case reports.

Materials and Methods: This study screened the published articles of case reports on PubMed from the American Journal of Orthodontics and Dentofacial Orthopedics (AJO-DO) dated from 1966 to 2021. AJO-DO was chosen because it is the official scientific journal of the American Association of Orthodontists (AAO). After being subjected to the inclusion and exclusion criteria, a total of 98 articles with 113 cases were included in the study.

Results: There is an increasing positive trend for reported records from 1966 to 2021. There is an increasing positive trend for report mandibular plane angle, lower incisor position, ideal overjet and overbite, class I cuspid relationship, class I molar relationship, and lip position. There is no change in the trend for arch form and intermolar distance.

Conclusions: From 1966 to 2021, there was an overall increasing trend in reported records according to the metrics that were examined in this study. The overall trend for reported outcomes was increasing as well from 1966 to 2021. The positive outcomes showed an increasing trend for the mandibular plane angle, lower incisor position, ideal overjet and overbite, class I cuspid relationship, class I molar relationship, and lip position. Arch form, intercanine distance, and intermolar distance were the least reported outcome. Ideal overjet and overbite were the most positively reported outcome measured. The lack of consistent reporting of all the metrics allows for continued improvement of case reports.

IMPACT OF THE COVID-19 PANDEMIC ON CAREGIVERS' ATTITUDES TOWARDS DENTAL CARE & PEDIATRIC ORAL HEALTH HABITS & DIET

Julia Geaumont, DMD; Arash Goshtasby, DMD; Maria A. Levi-Minzi, PhD; Jennifer Chung, PhD; Oscar Padilla, DDS

Purpose: This cross-sectional study aimed to determine the impact of COVID-19 on pediatric dental care, diet, and oral health habits.

Methods: Caregivers of pediatric patients (N=295) were surveyed on the child's diet, the child's oral health habits, and their attitudes toward attendance at dental appointments during the COVID-19 pandemic. Bivariate logistic regression was conducted to predict the outcome variable: caregiver willingness to take their child to the dentist during the lockdown, and significant variables ($p<0.05$) were added to a multiple logistic regression model.

Results: It was reported that 21.0% of patients decreased their brushing habits and 26.1% of caregivers increased their attention to their child's oral health. 31.9% of caregivers believed dental clinics were more dangerous for COVID-19 transmission than public spaces. Multivariate analysis found that willingness to take the child to the dentist was significantly higher among: patients with lower cariogenic diet before pandemic (OR: 2.42 [95% CI 1.37, 4.29], P=0.00), patients needing preventive care appointments (OR: 4.61 [95% CI 2.55, 8.32], P=<0.01), and caregivers that attended their own dental appointment (OR: 2.17 [95% CI 1.15, 4.13], P=0.02). Caregivers concerned about contracting COVID-19 at dental clinics had significantly lower odds
(OR: 0.30 [95% CI 0.15, 0.60], P=<0.00) of taking their child to appointments.

Conclusions: COVID-19 negatively impacted oral hygiene habits, diet, and attendance at dental appointments; resulting in an increase in caries risk of pediatric populations.

3D ANALYSIS OF IMPACTED CANINE POSITION IN VARIOUS SKELETAL RELATIONSHIPS: A RETROSPECTIVE STUDY

Hirsch, M; Choi, E.; Premaraj, S.; Hardigan, P.

Objectives: The objective of this retrospective study was to examine if there is an association between the maxillary impacted canine position in all three dimensions and different horizontal and vertical skeletal relationships.

Methods: This investigation was a retrospective study that examined the position of impacted canines in three dimensions using CBCT imaging. A sample size of 80 participants were used that were between 13 and 40 years old with no specific gender requirements. The sample size was divided in to three groups (class I, class II, class III) based on horizontal skeletal relationship and then into another three groups (normo-, hypo-, hyper- divergent) based on vertical skeletal relationship. Using the Dolphin Imaging Software, the CBCT DICOM files were uploaded so that linear, angular, and categorical measurements of the canine crown and root position were recorded digitally. Cephalometric radiographs were constructed from CBCT imaging and horizontal and vertical skeletal measurements were made digitally as well measured in millimeters and degrees. A one-way ANOVA test was used for the difference in means between groups and a Tukey HSD test was used for post model comparisons.

Results: No significant differences were found between maxillary canine position and horizontal skeletal relationships. A significant difference with a p-value of 0.01 from a one-way ANOVA test was found between vertical skeletal relationships and the mesio-distal rotation of the impacted canine in the axial view. A significant difference with a p-value of 0.02 from a one-way ANOVA test was found between vertical skeletal relationships and crown position of the impacted canine in the axial view. Tukey HSD test showed that hyperdivergent subjects had a significantly higher mean than normodivergent and hypodivergent subjects when comparing vertical skeletal relationship to crown position and mesio-distal rotation in the axial view. Hyperdivergent subjects had maxillary impacted canines significantly further from the mid-palatal suture and angulated away from the mid-palatal suture compared to other vertical skeletal relationships.

Conclusions: Maxillary impacted canine position was not significantly associated with horizontal skeletal relationship in this study, but did show a significant relationship when comparing vertical skeletal relationships in hyperdivergent subjects. Knowing maxillary impacted canines in hyperdivergent subjects are impacted further from the midline and better angulated, impacts treatment difficulty, treatment duration, overall treatment planning and the biomechanics needed to bring the canine into the arch.

COMMUNITY-BASED DENTAL CLINICS IN FLORIDA HEALTH DISPARITIES: COMPARISON OF PROCEDURES PROVIDED BY PEDIATRIC MEDICAID AND FREE SERVICES

Chung, J., Levi-Minzi, M., Ocanto, R., Rivas, Y., Padilla, O., Kodish Stav, J., Oramas, V.

Minority and low-income children in Florida are at the highest risk for oral health disparities, low health literacy, and difficulty accessing care. When compared with other racial and ethnic groups, non-Hispanic black Head Start children (28.9%) had the highest incidence of untreated decay (Florida Department of Health Public Health Dental Program 2021). There was also a direct relationship found between receiving free or reduced lunch at school (used as a measure of poverty status) and poor oral health outcomes; as free or reduced lunch percentage increased, the percentage of untreated decay increased from 5.3%-30%; caries experience increased (24.2% to 52.2%; early need for dental care increased (2.4% to 23.8%); and, urgent need for dental care increased from 1.3% to 4.4% (Florida Department of Health Public Health Dental Program 2021). Access to oral health services often relies heavily on the use of dental insurance. Head Start children without dental insurance experienced the highest levels of untreated tooth decay (28.5%) and caries experience (38.4%). These findings were similar among 3rd grade children in Florida, with 32.8% of those without insurance reporting untreated tooth decay.

Objective: To compare two Medicaid dental clinics and one free community based dental clinic in South Florida.

Methods: Treatment codes were extracted from aXium software for the time period of July 2017 through June 2022. Data from two Medicaid clinics were combined and compared to data from a free community based dental clinic providing services to children regardless of ability to pay. Descriptive statistics are reported.

Results: A total of 79,740 treatment codes were generated for the Medicaid clinics, and the free clinic yielded 15,744. The Medicaid clinics applied topical fluoride varnish most often (13.01%), followed by oral hygiene instruction (12.47%), periodic oral evaluation (10.57%), prophy (7.66%), and sealant application (7.37%). The free clinic performed sealant application most frequently (10.68%), followed by bitewing x-ray, (10.08%), topical fluoride varnish (9.22%), and oral hygiene instruction (9.12%); SDF application occurred at a significantly higher rate at the free clinic (7.27% versus 1.90% at the Medicaid clinics). In terms of restorative services, the Medicaid clinics provided more fillings (3.42% versus 2.87%), while the free clinic provided more extractions (2.38% versus 1.95%) and primary tooth crowns (3.11% versus 1.51%). Nitrous oxide was also more frequently used in the free clinic (5.36% versus 2.21%).

Conclusions: Untreated decay and dental caries remain serious health issues among Head Start children as the prevalence of these conditions has increased by 15.4% and 6.9% respectively from 2014-2015 to 2017-2018 (Florida Department of Health Public Health Dental Program 2021). Results indicate that both the Medicaid and free clinics provided preventive services most often (varnish and sealant application), however, it appears that the free clinic has more complex cases as evidenced by higher incidence of SDF application, fillings, extractions, crowns, and use of nitrous oxide.

PARENTS AND/OR GUARDIANS' SATISFACTION TOWARDS THE USE OF SILVER DIAMINE FLUORIDE AS AN ALTERNATIVE TREATMENT APPROACH TO CARIOUS LESIONS AFTER APPLICATION

Carly Cotten, DMD, MS; Jennifer Chung, PhD; Maria A. Levi-Minzi, PhD;
Romer Ocanto, DDS, MS; Marilyn Taylor, BA

Purpose: The goal of this study is to determine the parents/guardian's satisfaction of silver diamine fluoride (SDF) after use.

Methods: This study utilized a non-randomized convenience sample. Parents/guardians were educated on the uses of SDF, its benefits, and its side effects before treatment was rendered. After receiving the SDF treatment, 82 parents/guardians were given a 24-question survey. The survey was offered in both English and Spanish and collected information using Likert scale responses (ranging from 1 to 5), yes/no answers and fill in the blank. Descriptive statistics and mean levels of SDF satisfaction were calculated; t-tests and regression analysis were also used to examine SDF satisfaction. Significance level was set at $p < .05$ for all comparisons.

Results: Mean SDF satisfaction score = 3.95 (Add Range: SD=0.89). Regression results indicated that patient characteristics, including gender, age, behavior in the dental office and whether the patient had SHCN did not significantly predict SDF. In the model to test the relationship between SDF treatment characteristics and SDF satisfaction, a thorough SDF explanation, a willingness to repeat treatment, and being shown an SDF example photo significantly predicted SDF satisfaction, $F_{(7, 74)} = 9.96$, $p = <.001$ [95% CI 2.04, 3.77]. Factors that played into choosing SDF treatment (i.e., child' behavior, location of the tooth, type of tooth, and wanting to avoid OR visit) did not significantly predict SDF satisfaction. Parent or guardian concern about the appearance of the child's tooth after application also did not significantly predict SDF satisfaction. T-test results revealed no significant differences in SDF satisfaction based on patient characteristics. In terms of treatment characteristics, those who had SDF more than a year ago had significantly higher SDF satisfaction scores (4.21 versus 3.81). In addition, those who had SDF on a posterior tooth had significantly higher mean SDF satisfaction scores (4.09 versus 3.64). Those who were explained SDF very well and those shown an example photo also had significantly higher mean SDF satisfaction scores (4.13 versus 2.92 and 4.07 versus 2.71, respectively).

Conclusions: The parents and/or guardians were pleased (4.03 vs 3) due to the use of visual aids given before the application with the appearance related to the use of treatment, likely because they knew what the treated sites would look like. Results illustrate that most parents and/or guardians were likely (69.5%) to choose SDF as a treatment option again for their child, suggesting that pediatric dentists should offer SDF as an option for treating carious lesions. Findings also highlight that SDF satisfaction is related to a thorough and detailed explanation of SDF treatment and its consequences prior to administration, thereby suggesting that pediatric dentists take extra time with parents and/or caregivers considering SDF treatment.

P.GINGIVALIS VESICLES POLARIZE MONOCYTE-LINAGE-COMMITMENT TO MACROPHAGE VS. OSTEOCLAST DIFFERENTIATION-STAGE DEPENDENTLY

Elizabeth Leon, Shin Nakamura, Satoru Shindo, Maria Rita Pastore, Tomoki Kumagai, Xiaozhe Han, Shengyuan Huang, Sunniva Ruiz, Alireza Heidari, Toshihisa Kawai

Objective: Outer membrane vesicles (OMV) produced by *Porphyromonas gingivalis* (*Pg*) is thought to function as nanosized vehicles to disseminate its virulent factors in periodontitis. However, the effects of OMVs on differentiation and function of macrophage/osteoclast linage cells remain unclear. We aimed to investigate whether *Pg*-OMVs affect the monocyte's linage-commitment, especially that to osteoclastogenesis (OC-genesis).

Methods: OMVs were isolated from *Pg*W83 using an ultra-centrifuge (100,000xg, 1 hour). *Pg*-OMVs (1 µg/mL) were applied to murine RAW264.7 monocyte-like cells which were primed with RANKL for various periods (0, 24 and 72 hours). After incubating with RANKL for 6 days total, TRAP staining, pit-formation assay, qPCR (*oc-stamp*, *dc-stamp*, *acp5*, *mmp9*, and *nfatc1*) and flow cytometry (macrophage markers, F4/80 and CD11b) were performed. ELISA was conducted to monitor TNF-α and CCL2 productions. For statistical analysis, one-way ANOVA with Tukey-Kramer's multiple comparison test with significance at $p<0.05$ was employed.

Results: OC-genesis was suppressed by the addition of *Pg*-OMVs to unprimed RAW264.7 cells, whereas *Pg*-OMVs promoted the OC-genesis of those primed with RANKL for 24h and 72h. More specifically, *Pg*-OMVs increased TRAP+cells (>3 nuclei) in 24h-RANKL-primed RAW264.7 cells along with the proportional upregulations of OC-genesis-related genes, including, *dc-stamp*, *oc-stamp*, and *mmp9*, while *Pg*-OMVs-exposure to 72h-RANKL-primed RAW264.7 cells further promoted the emergence of large TRAP+cell (>10 nuclei) in conjunction with elevated *oc-stamp* expression, suggesting that *Pg*-OMVs promote OC-genesis in a manner dependent on OC-differentiation stage. *Pg*-OMVs increased CCL2 production more from the 72h-RANKL-primed RAW264.7 cells than those unprimed or primed with RANKL for 24h. According to flow cytometry, *Pg*-OMVs increased expression of the macrophage markers F4/80 and CD11b in unprimed RAW264.7 cells which were rather decreased in RANKL-primed RAW264.7 cells.

Conclusion: These results indicate that *Pg*-OMVs can polarize the monocyte-differentiation in a manner dependent on the monocyte-lineage-commitment stage for macrophage-genesis in early stage and for OC-genesis later, during inflammatory bone resorption process.

EFFECT OF SILVER DIAMINE FLUORIDE ON LACTOBACILLUS ACIDOPHILUS ASSOCIATED WITH EARLY CHILDHOOD CARIES.

Singana, T.; Chin, J.; Heidari, A.; Kawai, T.; Hardigan, P.

Objective: The purpose of this study was to evaluate the effect of SDF on the growth of Lactobacillus acidophilus and compare it to that of fluoride varnish and Chlorhexidine and to determine the minimum inhibitory concentration (MIC) of SDF on the growth of L.acidophilus

Materials and Methods: The antibacterial effect of the tested groups was determined using both disk diffusion assay and minimum inhibitory concentration (MIC) assay. In the disc diffusion assay, SDF (38%), fluoride varnish (5%), Chlorhexidine (0.12%, positive control) and PBS (negative control) were applied to a paper disc placed on the deMan, Rogosa, and Sharpe Agar (MRSA) plate on which low concentration (invisible level) of L.acidophilus was evenly spread (1×10^6/ml). After incubation at 37°C in anaerobic condition for 24 hours, the diameter of the inhibition zone around each disc was measured with a digital calliper.
In MIC assay, the minimum inhibitory concentration of SDF (38%) and Chlorhexidine (0.12%, positive control group) were tested by the exposure of L.acidophilus of 1×10^6 concentration in a 96-well plate to the serial dilutions of the respective reagent. After incubation at 37°C in anaerobic condition for 24 hours, the relative amount of L.acidophilus grown in the 96-well plate was measured using a plate reader and the minimum concentration of reagent that inhibited the growth of L.acidophilus was determined.
In the disc diffusion assay, the experiment was performed in quadruplicates and repeated three times. A general linear model (ANOVA) was employed for statistical analysis. In MIC assay, the minimum inhibitory concentration percentage of each material's original concentration was calculated.

Results: SDF (38%) demonstrated a significantly higher inhibitory effect ($P < .001$) against L.acidophilus   growth compared to Chlorhexidine. However, fluoride varnish showed slight inhibitory effect and PBS showed no inhibitory effect on the growth of L.acidophilus .
MIC of SDF was 0.006% of its original concentration, compared to 0.0003% of that for Chlorhexidine suggesting that chlorhexidine has more effect even at lower concentration than antibacterial effect mediated by SDF.

Conclusion: This study, for the first time, demonstrated the inhibitory effect of SDF on the growth of L.acidophilus, indicating that SDF possesses antibacterial property, which would be translated into the novel approach in arresting ECC associated with L.acidophilus .
This study allows a better understanding of the SDF effect in arresting ECC and discovers a new beneficial mechanism of SDF, in addition to its known mechanisms.

PATIENT ACCEPTANCE OF SCREENING FOR HEART DISEASE IN THE DENTAL CARE SETTING: RESULTS FROM A UNIVERSITY-SPONSORED DENTAL CLINIC

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Objective: Dental care settings provide potential opportunities for identifying patients at risk for heart disease who may not otherwise be detected. Our study examined correlates of patient acceptance of heart disease screening in a dental care setting.

Methods: We assessed patient characteristics, attitudes, beliefs, and behaviors from a cross-sectional survey of patients attending a university-sponsored dental clinic from 2010-2011 (n=400; 80% response rate). A multivariable logistic regression model examined the correlates associated with acceptance of heart disease screening.

Results: Almost all patients (95.2%) reported likely acceptance of screening for heart disease in the dental care setting if offered by their dentist. Patients who were aware of a relationship between oral and overall health, versus those not aware, as well as patients who reported a moderate or high risk of heart disease, versus those with no or low risk, demonstrated the highest adjusted odds ratios (AOR) of likely acceptance of heart disease screening (AOR = 5.11, 95% CI 1.72, 15.21, and AOR = 3.50, 95% CI 1.21, 10.15, respectively).

Conclusions: We report a high prevalence of acceptance of heart disease screening among patients attending a university-sponsored dental clinic. Our study highlights the associations of patient awareness of the relationship of oral health to general health and self-rated risk of heart disease, with acceptance of heart disease screening the dental care setting.

Practical Implications: Patient acceptance of heart disease screening in the dental care setting provides a potential public health opportunity for early identification of heart disease risk and disease prevention.

Human Participant Protection: The institutional review boards at the University of Miami and Nova Southeastern University approved this study. University of Miami IRB Number: 20090802

THE BACTERIOLOGY OF SUPRAGINGIVAL PLAQUE OF ADULTS IN SOUTH FLORIDA: TRENDS IN COLONIZATION

Demehri, S.; Ozga, AT; Vardar-Sengul S; Garcia-Godoy, Cristina

Objectives: To investigate the oral microbiome of adults in South Florida in order to understand the impact of demography and sampling location on overall bacterial abundance and diversity.

Methods: A total of 27 patients, ages 18-38 were initially screened and their demographic information, medical history, and periodontal health were assessed at the time of informed consent. A tongue scrape and supragingival plaque samples from each quadrant of the mouth were collected using sterile curettes. Each quadrant and tongue samples were frozen separately in Tris-EDTA Buffer. The plaque, stored in a Tris-EDTA buffer, underwent extraction using the Qiagen BIOstatic Bacterermia DNA Kit, and the V4 hypervariable region of the bacterial 16S ribosomal rRNA gene was amplified for taxonomic purposes. PCRs were completed in duplicate and pooled in equimolar amounts, purified, and sequenced using an Illumina MiSeq (V3 chemistry) sequencing platform. Samples were assessed for overall abundance and rarefied to 10,000 reads for diversity analyses within QIIME2 and results were further analyzed using packages within R.

Results: A total of 8,698,149 de-multiplexed reads were recovered from 5 oral sites across 27 patients. After <Q30 reads were removed, a total of 7,418,279 reads were used for downstream analyses. Samples were dominated by Firmicutes (4.7-75%), Bacteroidota (1.1-54.5%), Proteobacteria (0.2-89.5%), and Actinobacteria (0.5%-67.2%) at the phyla level along with Streptococcus (0.7-50.1%), Veillonella (0-37.5%), and Prevotella (0-42%) at the genus level (Figure 1). Total observed OTUs (alpha diversity- Figure 2) and weighted unifrac values (beta diversity) differed significantly based on oral sampling site and gingivitis assessment. Although alpha and beta diversity differed between participants the main drivers of the differences across oral taxa is the location of sampling within the mouth (tongue being an outlier) and oral health assessment (gingivitis vs. healthy).

Conclusions: Overall, the results show a range of common commensal and pathogenic microbiota within adults from South Florida. This research stresses the need for standardized sampling for oral microbiome studies, a thorough examination of patient health states, and the inclusion of individuals from underrepresented groups in future examinations.
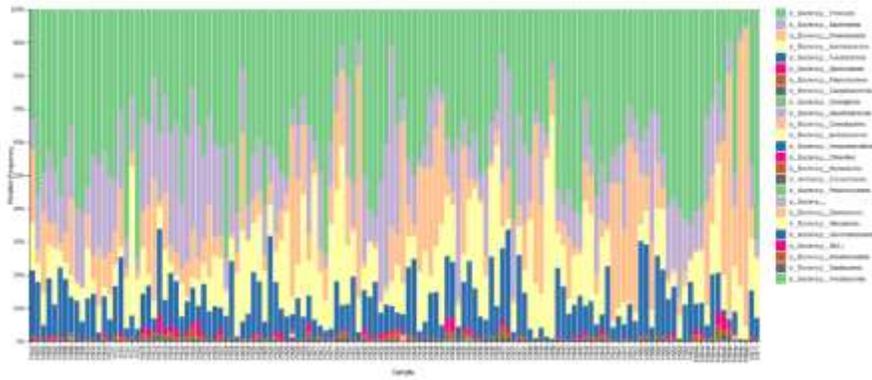
Figure 1: Abundance profiles at the phyla level organized by participant

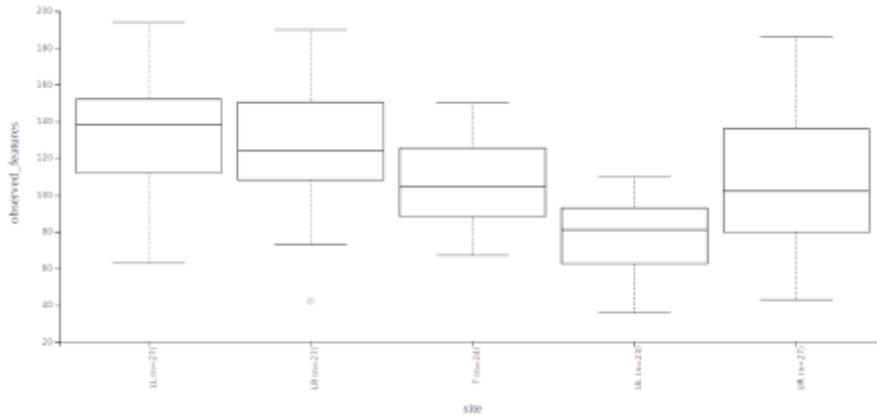**Figure 2: Alpha diversity of each oral site based on observed species**

OC-STAMP-REACTIVE IGG AUTO-ANTIBODY UPREGULATES OSTEOCLASTOGENESIS IN ASSOCIATION WITH ELEVATED ANTI-*P. GINGIVALIS* ANTIBODY

Shin Nakamura, Satoru Shindo, Elizabeth Leon, Xiaozhe Han, Tomoki Kumagai, Maria Rita Pastore, Alireza Heidari, Kazuhisa Ouhara, Maria Hernandez, Saynur Vardar-Sengul, Toshihisa Kawai.

Objectives: Periodontal disease (PD) is a polymicrobial inflammatory lesion in which pathogenic bone loss is caused by osteoclasts (OC) differentiated from MCSF/RANKL-primed monocytes. Elevated IgG antibody to a keystone opportunistic pathogen, *Porphyromonas gingivalis* (*Pg*), is a hallmark humoral response in PD that is correlated with pathogenic bone loss. On the other hand, osteoclast stimulatory transmembrane protein (OC-STAMP) is a unique cell membrane protein which is distinctively expressed on OC precursors and monocytes. Interestingly, while OC-STAMP is engaged in cell-fusion event required for *in vitro* OC-genesis, its role in pathophysiological context of periodontitis and other bone lytic diseases remains elusive. We previously reported that OC-STAMP promoted a pathogenic bone resorption using mouse ligature-induced PD model. This study examined the humoral immune responses to OC-STAMP expressed by human monocytes in relation to anti-*Pg* antibody.

Methods: Blood serum (N=40) collected from systemically healthy human subjects (Precision for Medicine, BocaBiolistics, and Innovative Research) were measured for IgG antibody titer to *Pg* or OC-STAMP peptides. Human peripheral blood mononuclear cells (PBMCs) were stimulated with MCSF/RANKL with or without anti-OC-STAMP rabbit polyclonal antibody or control rabbit IgG. Affinity purified human anti-OC-STAMP antibody from serum with elevated anti-*Pg* antibody titer was also tested for its activity to alter the OC-genesis. OC-genesis induced *in vitro* was evaluated by TRAP staining and pit formation assay. Western blot was performed to confirm whether anti-OC-STAMP antibody in human serum reacts to OC. Statistical differences were assessed using one-way ANOVA with Tukey-Kramer's multiple comparison test. The correlation between IgG antibody titer against OC-STAMP and anti-*Pg* antibody titer was evaluated using the Pearson product-moment correlation. *P* value <0.05 was considered statistically significant.

Results: IgG antibody titer to *Pg* positively correlated with that to human OC-STAMP peptides (r=0.578, *p*<0.01). Anti-OC-STAMP auto-antibody purified from human serum with high IgG antibody titer to *Pg*, as well as anti-OC-STAMP rabbit antibody significantly promoted *in vitro* OC-genesis (*p*<0.05). Anti-OC-STAMP antibody in human serum with elevated anti-*Pg* antibody titer bound to OC-STAMP from osteoclasts lysate.

Conclusions: The results indicated that *Pg* infection may be associated with development of anti-OC-STAMP auto-antibody which can promote alveolar bone destruction via upregulation of OC-genesis, suggesting a novel immunological intervention in the pathogenesis of PD.

THE EFFECTS OF INTRAOSSEOUS DEPTHS AND EXTRAOSSEOUS LENGTHS ON IMPLANT STABILITY
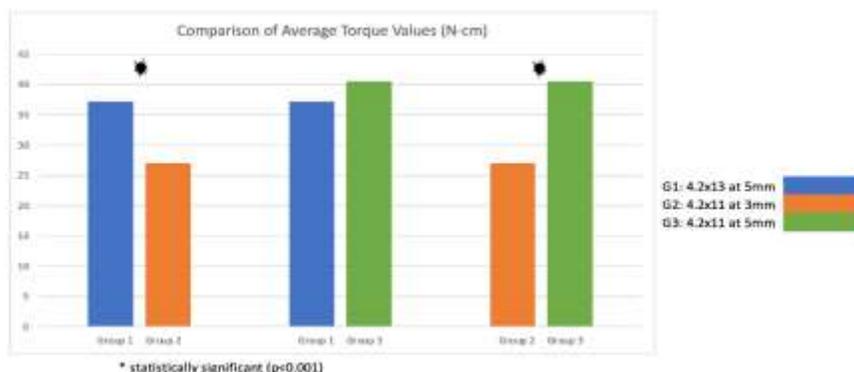
Hatara, A., Koutouzis, T., Sofos, S.

Introduction: Immediate implant placement is a routine procedure and the decision whether or not to immediately load is often based on implant stability. Stability can be quantified using several methods, most common being insertion torque values (N-cm) and resonance frequency analysis values (RFA).
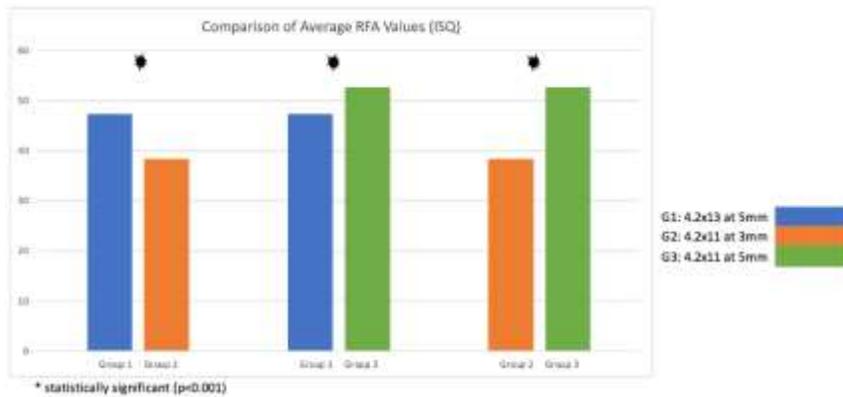
Objectives: To compare insertion torque values (N-cm) and resonance frequency analysis values (RFA) in simulated immediate implants placed at different intraosseous depths with identical extraosseous lengths; and in simulated immediate implants placed at different extraosseous lengths with identical intraosseous depths.

Methods: 90 Dentsply Sirona PrimeTaper implants were divided into 3 groups. Group 1 consisted of 30 4.2x13 mm implants, while Groups 2 and 3 consisted of 30 4.2x11 mm implants each. Using the PrimeTaper drilling protocol, osteotomies were created in a solid rigid polyurethane foam block which mimics D2/D3 bone quality, and at a 45 degree angle to a depth of 5 mm for Groups 1 and 3, and 3 mm depth for Group 2. Implants were placed until the minimum intraosseous depth was 5 mm for Groups 1 and 3, and 3 mm intraosseous depth for Group 2, simulating an immediate implant in a fresh extraction socket. Insertion torque measurements were taken with the supplied Denstly Sirona Torque Wrench EV while RFA measurements were taken at 2 points using the Osstell device. For description of the mean data values, standard deviations and two way analysis variance (ANOVA) with Bonferroni post hoc were calculated.

Results: Statistically significant differences were found when comparing average torque values between Group 1 vs Group 2, and Group 2 vs Group 3 (p<0.001). There was no statistically significant difference when comparing Group 1 vs Group 3 torque values. The average torque values for Groups 1, 2, and 3 were 37.17 N-cm, 27.00 N-cm, and 40.50 N-cm respectively.

Statistically significant differences were found when comparing average RFA values between Group 1 vs Group 2, Group 1 vs Group 3, and Group 2 vs Group 3 (p<0.001). The average RFA values for Groups 1, 2, and 3 were 47.35, 38.33, and 52.66 respectively.

Comparison of Average Torque Values (N-cm)

G1: 4.2x13 at 5mm
G2: 4.2x11 at 3mm
G3: 4.2x11 at 5mm

* statistically significant (p<0.001)

Comparison of Average RFA Values (ISQ)

G1: 4.2x13 at 5mm
G2: 4.2x11 at 3mm
G3: 4.2x11 at 5mm

* statistically significant (p<0.001)

Conclusions: With respect to torque values, the tested implant groups showed that the intraosseous depth is significant. It seems reasonable to infer that as long as the intraosseous depth is similar, the extraosseous length does not influence the final implant insertion torque.

The test groups further showed that RFA values are influenced by both the extraosseous implant length as shown by the significant difference between Groups 1 and Group 3 and also by the intraosseous depth as shown by significant difference between Group 1 and Group 2.

Clinical Significance: Traditionally, when loading immediate implants, insertion torque was considered an important factor in determining implant stability. Furthermore, this study has shown that the extraosseous length has an effect on RFA values therefore the decision of immediate loading should take into consideration the extraosseous length.

Support: This study was supported by Dentsply Sirona.

THE ROLE OF THE ORAL MICROBIOME IN SARS-COV-2 INFECTION

Bontempo A; Chirino A; Arora S; Bomparai S and Cayabyab MJ

Objectives: The devasting COVID-19 pandemic has claimed the lives of over 5 million people globally. COVID-19 is caused by a novel Beta coronavirus, Severe Acute Respiratory Syndrome Corona Virus 2 (SARS-CoV-2) and the virus establishes infection mainly in mucosal epithelial cells found in the nose and lungs. The oral mucosa is also proposed to play an important role in COVID-19 infection since the cells in the mouth were shown to be infected. Because the oral cavity is an important site of infection, we explored the role of the oral cavity, specifically the role of the oral microbiome in COVID-19 infection since previous studies suggest that the oral microbiome is likely to play an important role in viral infection. We observed that the oral bacteria isolated from 37 human saliva samples were capable of either promoting or inhibiting Cov-2 infection in vitro. Furthermore, we found that certain bacterial species (i.e *Porphyromonas gingivalis*) can inhibit infection. We are currently identifying factors produced by oral bacteria that can modulate COVID-19 infection. The mechanisms by which oral bacterial factors can mediate either enhancement or inhibitions will be discussed. These studies underscore the importance of the oral microbiome in SARS-Cov-2 infection and transmission.

Methods: We developed a CoV-2 and VSV-g pseudotyped dual reporter (Luc/GFP) lentiviruses based on a second-generation design using pCMVdR8.2 backbone plasmid, along with stable ACE2+ 293T cells. This model permits us to analyze the effect of oral factors in the virus entry step. To assess Pg factors in CoV-2 infection, Pg supernatant were tested for antiviral activity. To assess oral effects on virus binding, virus was loaded to the BLI biosensors using a ACE2-Fc tagged previously bound to HFC biosensors and analyzed with a Gator BLI equipment. We established a Syncytia model to assess the viral entry step at which the inhibition was occurring. To Lastly, to assess the effects of the oral microbiome in CoV-2 infection, the oral bacteria in the human saliva were cultured in TSB overnight then tested for anti-Cov-2 activity.

Results: Pg supernatant and PGDHC showed strong virus inhibition. Post-entry and syncytia formation assay indicate the effect is at the entry level, although it is not by interfering with ACE2/spike interaction as suggested by BLI data. KGP and RGPA, but not RGPB enhanced infection by approximately 50%. This effect is not observed against VSV-g pseudotyped lentivirus. Further isolation and characterization of the active compound are in process. The analysis of 37 human saliva microbiome showed both enhancing/inhibitory among the population, suggesting a relevant role for the oral microbiome in CoV-2 infection.

Conclusion: In conclusion, our data support the role of oral microbiome in Covid-19 infection and warrant an in-depth analysis of the mechanisms involved in oral microbiome, host and CoV-2 viral interactions. In addition, our observations could be extended to other oral viral infections including Hepatitis, EBV, influenza.

COMPARING STRAIGHT VERSUS ANGLED SCREW CHANNEL SYSTEMS IN ANTERIOR MAXILLA USING CBCT FOR PLANNING IMMEDIATE IMPLANT PLACEMENT: A RETROSPECTIVE STUDY

Muhammad Taimur Khan, Niranjan Ratnakar Joshi.

Objectives: Immediate implant placement in the esthetic zone is technique-sensitive and requires careful assessment of factors such as surgical technique, anatomic considerations and prosthetic planning. This is critical to ensure immediate implant placement in the correct three dimensional (3D) position with an intact facial bone wall to obtain ideal, predictable and stable esthetic outcomes. The use of cone beam computed tomography (CBCT) is the gold standard for providing three dimensional (3D) images for planning dental implants. The aim of the study is to assess the possibility of screw retained implant restoration using straight versus angled screw channel systems in the anterior maxilla during implant planning on CBCT scans.

Methods: The CBCT scans of patients who were treated at the Postgraduate dental clinics at Nova Southeastern University from 2017-2023 were selected upon IRB approval, and CBCT scans were imported in implant planning software for planning Astra EV implants in the anterior maxilla using straight access and angled screw channel abutments. This was done using a template (Figure.1) on the implant planning software to measure the angulation needed to achieve a screw- retained restoration for each tooth type. The angulation was incrementally increased from 5 degrees to 25 degrees until the access channel is exiting palatally and with 2 mm distance from the incisal edge. All the abutments planned within this range were deemed restorable with an angled screw channel. If a 25 degree angled abutment is still exiting facial to the incisal edge or not 2 mm palatal to it then they will be deemed restorable with a cement retained restoration. The prevalence of each restoration type was then compared among the tooth types,i.e. centrals, laterals and canines by using Pearson chi-square test. Statistical significance was evaluated within these groups for the restoration types to achieve a screw retained restoration.
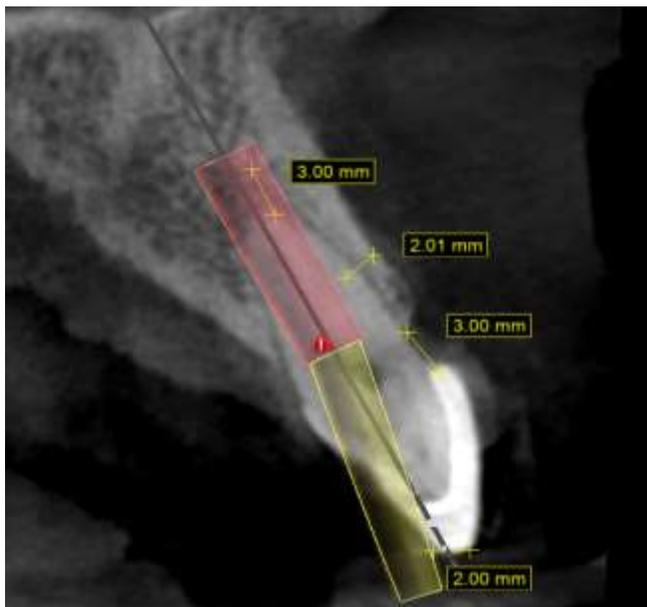
Figure. 1

Results: A total of 100 CBCT scans were evaluated that involved 110 sites in the anterior maxilla. The results demonstrated that 85 sites (78%) required an angled screw channel system to obtain a screw retained restoration on immediate implant placement. On the other hand, 32 sites (22%) required straight abutments only for a screw retained restoration. There was a significant increase in screw retained restorations on using angled screw channel systems ($P<0.05$). The lateral incisor was the tooth that required the most significant angle correction among the anterior teeth. The required angle correction was 20 degrees that allowed screw retained restorations more frequently (78%) within the sample.

Conclusion: The study showed that the angled screw channel systems significantly ($p<0.05$) increased the prevalence of a screw retained restorations in the anterior maxilla. Lateral incisors required the greatest amount of angle correction within the group.

P. GINGIVALIS OMV-INDUCED APOPTOSIS, PD-L1 EXPRESSION AND FUNCTIONAL CHANGES IN THP-1-DIFFERENTIATED MACROPHAGES

**Islam Saleh, Shengyuan Huang, Sunniva Ruiz, Xiaozhe Han.**

Objectives: Microbial outer membrane vesicles (OMVs) have been suggested to play an important role in the host immune modulation and inflammatory responses. This study is to determine the effect of *P. gingivalis* OMVs on apoptosis and programmed death-ligand 1 (PD-L1) expression of THP-1-differentiated macrophages.

Methods: The supernatant of *P. gingivalis* (ATCC 33277) culture was ultracentrifuged at 100,000g for 2 h at 4C° to isolate OMV, which was directly quantified using nanoparticle tracking analysis (NanoSight NS300). THP-1 cells were cultured in RPMI 1640 Medium with 10% FBS and 1% antibiotic-antimycotic mix. The following different phenotypes of THP-1-differentiated macrophages were obtained: M0 (100nM PMA, 24h); M1(2µg/ml *P. gingivalis* LPS + 20ng/ml IFN-γ, 48h); M2 (20ng/ml IL-4 + 20ng/ml IL-13, 48h). Differentiated macrophages were treated with 50µg/ml *P. gingivalis* OMV for 24h and then harvested for apoptosis staining with Violet ratiometric membrane asymmetry probe with dead cell discrimination kit followed by flow cytometry. Programmed death-ligand 1 (PD-L1) expression in macrophages was detected by mouse anti-human PD-L1 AF488 antibody followed by flow cytometry. Macrophage phagocytosis activity and polarization were detected by florescent microscopy and flow cytometry.

Results: The percentage of apoptotic cells in cultured M0 macrophages were not changed in *P. gingivalis* OMV treated group compared to control group. However, significantly increased apoptotic induction were observed in *P. gingivalis* OMV-treated M1 macrophages (14.07% vs. 3.13%) and M2 macrophages (49.13% vs. 6.33%). A significant increase of PD-L1 expression was observed in M0 and M2 macrophages after *P. gingivalis* OMV stimulation. The PD-L1 expression by M1 macrophages was not significantly changed in the presence of *P. gingivalis* OMV compared to control. Phagocytosis activity in M0 (5.56% vs. 11.82%) and M2 macrophages (7.14% vs. 14.25%) was enhanced by *P. gingivalis* OMV treatment, but was not changed in M1 macrophages by *P. gingivalis* OMV treatment since these cells already bear high level of phagocytotic activity. Surprisingly, M2 macrophage underwent significant phenotype switch to M1 as indicated by the substantial increase in M1 population and decrease in M2 population.

Conclusion: *P. gingivalis* OMVs drive both naive and polarized macrophages mainly towards pro-inflammatory M1 phenotype differentiation by differentially modulating cell turnover, immune check point engagement, and phagocytosis function of M0/M1/M2 macrophages, which may potentially disrupt the balanced host immune and inflammatory responses.
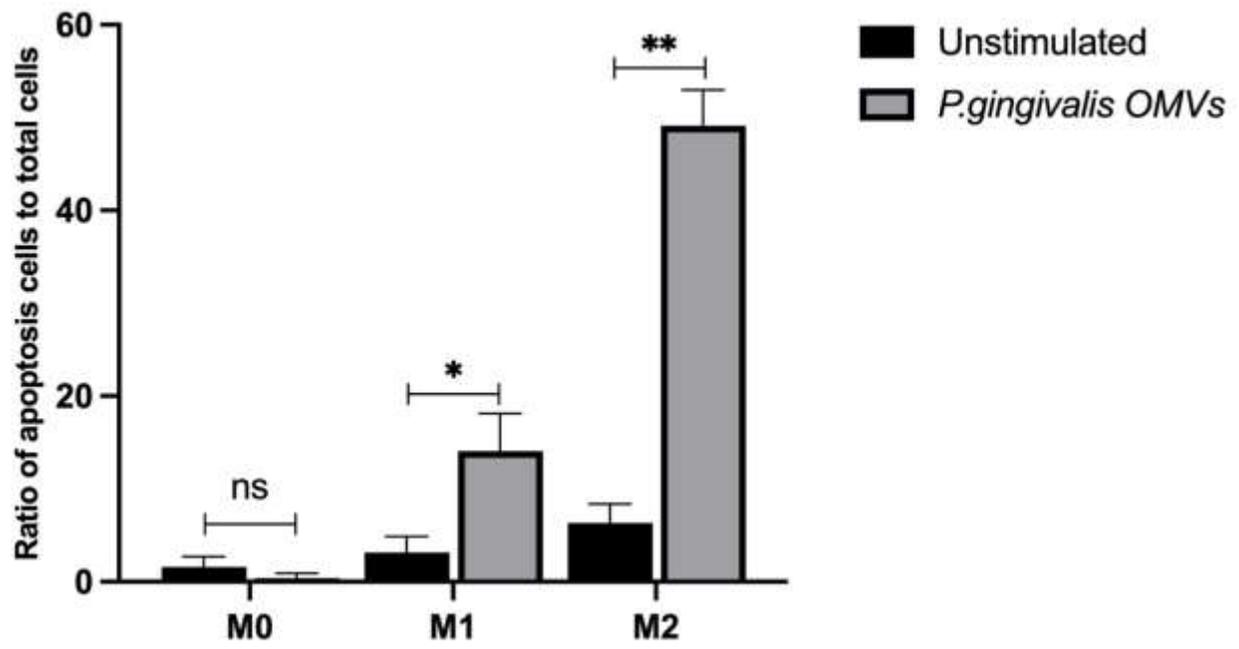
SLEEP-RELATED BREATHING DISORDERS AND PERIODONTAL CONDITION: A STUDY USING CONE-BEAM COMPUTED TOMOGRAPHY OROPHARYNGEAL VOLUMETRIC AIRWAY ANALYSIS

Khan, S., Cummings, N., Vardar-Sengul, S., Bozzutti, G.

Objectives: To evaluate if severity of periodontal disease (PD) and/or radiographic signs of parafunction, including periodontal ligament widening, cuspal fracture, and tooth wear, are associated with the presence of a sleep-related breathing disorder and/or oropharyngeal airway volume as measured by cone-beam computed tomography (CBCT).

Methods:

Male and female adult patients in a university faculty practice setting, treated by board-certified periodontists, were cross-sectionally analyzed for periodontal data, medical histories, and cone-beam computed tomography interpretation. Patients with a diagnosis of periodontitis were staged and graded according to the 2017 Classification of Periodontal Diseases and Conditions. Medical histories were evaluated for patient-reported snoring and/or a previous diagnosis of obstructive sleep apnea (OSA). Oropharyngeal airway volume (mm^3) was measured by cone-beam computed tomography. Confounder variables were recorded in patient's medical histories including: diabetes mellitus (type 1 or 2), hypertension, coronary artery disease, hypercholesterolemia or hyperlipidemia, chronic obstructive pulmonary disease, nasal stuffiness, tobacco use, and alcohol use. Descriptive statistics, and inferential statistics, including bivariate analysis and nominal regression, were conducted via Pearson Chi-square tests of statistical significance. Risk estimation via odds ratio was performed for sleep-related breathing disorders and patient periodontal clinical traits involving radiographic signs of parafunction.

Results:

Thirty-six patients diagnosed with absence of PD or gingivitis or Stage 1-2 periodontitis, Stage 3 periodontitis, and Stage 4 periodontitis at frequencies of 25.0%, 41.7%, and 33.3%, respectively. Twenty-two patients were analyzed for statistical analysis of oropharynx volume, ranging in value from 2 mm^3 to 19 mm^3, with a mean of 7.7 mm^3. A non-statistically significant interaction was found between snoring and severity of PD (X^2 (2) = 0.059, p = 0.971), as well as between a diagnosis of OSA and severity of PD (X^2 (2) = 0.741, p = 0.690). A non-statistically significant interaction was found between oropharyngeal airway volume and PD severity (X^2 (40) = 45.6, p = 0.251). A non-statistically significant association was found between the exposure of snoring and radiographic signs of parafunction (OR = 2.91, 95% CI: 0.597-14.185), as well as between OSA and radiographic signs of parafunction (X^2 (1) = 1.895, p = 0.169).

Conclusions:

PD and sleep-disordered breathing have been studied with respect to a bidirectional, unidirectional, or no relationship association, yielding mixed evidence. The current study demonstrated no significant association between periodontitis and sleep-disordered breathing, either by patient-reported snoring or previous diagnosis of OSA, as well as no significant

association between periodontitis and oropharyngeal airway volume measured by CBCT. No significant association was found between oropharyngeal volume as measured by CBCT and sleep-disordered breathing. Based on the results of the current pilot study with a limited sample size, it can be concluded that further association studies with larger sample sizes must be conducted to discern the nature of the relationship between sleep-related breathing disorders and PD.

VITAMIN D INFLUENCE ON IMPLANT OSSEOINTEGRATION.
A SYSTEMATIC REVIEW AND META-ANALYSIS.
Wang A, Garcia M.

Background: It is imperative to understand the systemic and local factors that allow for dental implant success as well their failures. Vitamin D deficiency may play an essential role in the development of implant failures.

Objective: This systematic review aimed to evaluate the current available evidence of the influence of vitamin D levels on dental implant osseointegration.

Material and methods: A focused question was constructed based on the PRISMA guidelines. The focus question was: "Does Vitamin D influence osseointegration around dental implants?" Searches were conducted through PubMed, EMBASE, and ResearchGate databases from the year 2008 up to 2022. A combination of the following keywords was used: 'Vitamin D', 'dental implant', 'osseointegration', 'implant failure'. Inclusion criteria were: animal and human studies in which dental implants were placed, studies that evaluated the association between vitamin D levels of subjects and osseointegration of implants, and clear outcome measures of osseointegration of implants. Exclusion criteria were letters to editors, case reports, reviews, and articles published in languages other than English. Success of dental implants was commonly defined by percent of bone-to-implant contact (%BIC). Vitamin D was assessed by serum levels of 25(OH)D or 25-hydroxyvitamin D. Vitamin D deficiency was defined as serum levels of 10 to 29 ng/mL and normal was serum levels greater than 30 ng/mL. Titles and abstracts of studies identified using the PRISMA protocol were screened by the authors (A.W. and M.G.) and checked for agreement. Full texts found to be relevant were read and independently evaluated by both authors for the above stated eligibility criteria.

Results: Seven experimental studies with animal subjects and five clinical studies with human subjects were included. Due to the differing outcome variables in human clinical studies, a meta-analysis was only performed on the animal experimental studies. There was a significant difference (p<0.05) in mean %BIC between a vitamin D deficient or enriched diet and a normal diet, and between vitamin D-treated implants and non-vitamin D-treated implants. Furthermore, there was a significant difference (p<0.05) in mean %BIC in a normal diet and vitamin D-treated implant versus normal diet and non-vitamin D-treated implant. Vitamin D deficiency had a negative impact on mean %BIC in animal studies. Two human studies failed to show a significant relationship between low serum levels of vitamin D and implant failure. One human study found that genetic polymorphism in the vitamin D receptor (VDR) gene may be a marker for implant loss, while another study stated other factors such as primary stability, surgical technique, and bone quality, but not VDR polymorphism were associated with implant loss.

Conclusion: Within the limitations of this study, vitamin D deficiency shows a negative influence on implant osseointegration in terms of mean percent bone-to-implant contact, There is limited

evidence that serum levels of vitamin D or vitamin D-treated implants influence implant osseointegration in humans.

Presentations
2023 June 7

EXPRESSION OF IL-1B IN ORAL EPITHELIAL CELLS (OBA-9), PERIODONTAL LIGAMENT CELLS (PDL), THP-1 CELL LINE (MONOCYTE), AND HUMAN GINGIVAL MESENCHYMAL STEM CELLS (HGMSCS) UPON EXPOSURE TO INVISALIGN® SMARTTRACK™ ELUTE: AN *IN VITRO* STUDY

Nachum,K., Premaraj,T. Heidari, A., Han,X.

Objective: To determine if there is an induction of inflammation in OBA-9, PDL, THP-1, and hGMSC cells upon exposure to Invisalign® SmartTrack™ material elute.

Methods: Invisalign® trays were soaked in Phosphate buffered (PBS) solution for a period of 3 days, 1 week, 2 weeks, and 4 weeks. The solution was then collected and added to *In-vitro* grown oral epithelial (OBA-9) cells, Periodontal Ligament (PDL) stem cells, THP-1 Cell Line (Monocyte), and Human Gingival Mesenchymal Stem Cells (hGMSCs) to form the Invisalign® elute. Each cell was cultured in its appropriate medium according to protocols. Invisalign® and PBS (control) were added at different concentrations to the cells. After 24 hours, the cell medium was tested for toxicity using water-soluble tetrazolium salt (WST-1) assay. Furthermore, pro-inflammatory cytokine release of Interleukin-1β (IL-1β) was profiled using enzyme- linked immunosorbent assay (ELISA). Each experiment was repeated three times and statistical analysis was done accordingly.

Results: Our findings demonstrated a time-dependent release of IL-1β upon exposure to Invisalign® elute to THP-1, OBA-9, and PDL cells, while no distinguishable pattern was determined for hGMSCs. The release of IL-1β was not statistically significant when compared to control (p>0.05). However, the WST-1 cell viability assay revealed that lower concentrations of the elute produced little to no cytotoxicity, while higher concentrations of elute resulted in moderate to severe cytotoxicity.

Conclusion: There was some evidence to suggest that SmartTrack™ material induces inflammation. Our results showed an increase release of IL-1β upon exposure to higher concentration of elute, however, no statistical significance was found. Monocytes, epithelial cells, and periodontal ligament cells showed an increased cell death upon exposure to higher concentration of SmartTrack™ elute, indicating cell cytotoxicity. Our study suggest that SmartTrack™ material induces cell death and promotes the release of inflammatory cytokine, IL-1β. However, interpreting *in-vitro* effects on patient care should be exercised cautiously. More research is needed to elucidate the mechanism of cell death and clinical effects on patients.

COMPARISONS OF RESISTANCE TO SLIDING OF AESTHETICALLY COATED AND UNCOATED ARCHWIRES AFTER LONG-TERM IMMERSION IN WATER

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Objectives: The objective of this study was to compare the resistance to sliding (RS) of aesthetically coated and uncoated orthodontic archwires in ceramic brackets at various bracket angulations after water immersion up to 24 weeks.

Background: Polymer-coated orthodontic wires have shown distinct material properties when compared to uncoated stainless-steel wires. Prolonged immersion in water, the most abundant component in human saliva, for up to 4 weeks has been shown to impact the resistance to sliding of polymer-coated wires, as a result of the plasticizing effects of water. While water is not a perfect substitute for oral saliva due to the absence of various organic and inorganic components, utilizing salivary substitutes would introduce additional variables which could hinder the study's objectives. Clinically, orthodontic wires could be used continuously for months. Therefore, the purpose of the current study was to compare RS of aesthetically coated and uncoated wires after water immersion for up to 24 weeks.

Materials and Methods: Three groups of 0.019 x 0.025 inch stainless-steel archwires were utilized: uncoated (Group U), epoxy-coated (Group E), and Teflon-coated (Group T). Each group was immersed for 0 weeks (T0), 8 weeks (T8), 16 weeks (T16), or 24 weeks (T24), in deionized water at 37°C. Maximum resistance to sliding (MRS) and average resistance to sliding (ARS) were measured by sliding the archwires through 0.022 inch slot ceramic brackets in either 0° (passive) or 3° (active) contact angle configurations using an Instron Universal testing machine. The data was then analyzed with a three-way ANOVA with independent variables of water immersion times, wire groups, and contact angles.

Results: All three independent variables played significant roles in RS. Pair-wise comparison of MRS and ARS at 3° contact angle showed that Group U was in the highest group and Group T was in the lowest for all timepoints. However, MRS and ARS at 0° contact angle showed distinct patterns different from that found in active configurations. Within wire-group comparisons of RS showed that the only statistically significant difference in MRS and ARS was found at a 3° contact angle in group E between T0 and T8.

Conclusions: There are distinct differences in the resistance to sliding of aesthetically coated and uncoated wires after long-term immersion in water for up to 24 weeks, with Teflon coating displaying the lowest MRS in active bracket configuration at all immersion timepoints, and uncoated archwires displaying the highest. Therefore, Teflon coated archwires may lend themselves readily to long-term sliding mechanics, whereas uncoated archwires are best utilized for anchorage and closing loop mechanics.

EFFECTS OF COFFEE AND OTHER BEVERAGES ON THE FORCE PROFILE OF ELASTIC POWER CHAINS OVER TIME; AN INVITRO STUDY

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Objectives: The effect of three common beverage choices on the force decay of orthodontic elastomeric power chains were evaluated.

Methods: In this in vitro study, 40 elastomeric chains of the same trademark were investigated. The experimental groups consisted of 3 different beverages: Hot Espresso Coffee, Coca-Cola soda, Gatorade Lemon Lime sports drink–plus a control group, which is Deionized Water. Force was measured at the following intervals: baseline (immediate exposure), 1 hour, 24 hours, 7 days, and 21 days. Tensile testing was executed using a Universal Testing Machine 8841 (Instron, Norwood, MA) with a crosshead speed of 20 mm/min. Descriptive statistics including the mean values and standard deviations of automatically recorded tensile loads were calculated for each sample at each time point. Data was analyzed by means of two piece HSD Test and one way ANOVA at a significance level of 0.05.

Results: Our analysis revealed significant differences between several beverage comparisons at the 0-hour mark (Significance at $P < 0.05$, Water-Gatorade, Water-Coca-Cola, and Coca-Cola-Coffee). However, no other significant differences were observed at this time point. Additionally, our analysis revealed significant differences between Coffee and every other beverage at 1 hour, 24 hours, and 7 days (Significance at $P < 0.05$, Coffee-Water, Coffee-Gatorade, Coffee-Coca-Cola). At the 21-day mark, we observed significant differences between all comparisons except for Gatorade-Coca-Cola (Significance at $P < 0.05$, Coffee-Water, Coffee-Gatorade, Coffee-Coca-Cola, Gatorade-Water, Coca-Cola-Water).

Conclusions: 1) At zero hour mark there was statistical significance of Water having more force decay than Coca-Cola, and Gatorade. Coffee had more force decay than Coca-Cola at the zero time point that was statistically significant as well. 2) At 1 Hour, 24 Hour, and 7 Days; Coffee had statistically significant force degradation of the elastics compared to Gatorade, Water, and Coca-Cola. 3) At 21 days, Coffee had the most statistically significant force degradation of the elastics over Gatorade, Coca-Cola, and Water. At the 21 day time point, Water showed statistically significant force decay of elastics when compared to Gatorade and Coca-Cola. At the 21 Day time point, Coca-Cola and Gatorade had no statistically significant difference in force decay of elastics.

SHEAR-BOND-STRENGTH OF SELF-ETCH OPTIBOND UNIVERSAL-360 TO VARIOUS DARK-CURED RESIN CEMENTS

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Introduction: It is critical to have reliable bonding systems to assure functional and esthetic longevity of indirect restorations. It is recommended to use resin cements with their respective bonding agents. However, this creates over-purchased products with expiration dates. Having a compatible universal adhesive is clinically desirable to optimize inventory control and improve product reliability issues with resin cements.

Objective: To comparatively evaluate the shear-bond-strength (SBS) of self-etched Optibond Universal 360 (OU360) when used with various dark-cured resin cements on dentin against its respective resin cement.

Methods: Fifty non-carious unidentified human posterior teeth were used after embedding in acrylic and exposing dentin with 400 followed by 600 grit size SiC paper, and ultrasonically cleaning for 10 min. The specimens were examined to assure the absence of caries and cracks under the optical microscope. They were divided in 5 groups of resin cements(n=10/group): Maxcem Elite(ME), RelayX Universal(RX), Calibra Ceram(CC), Variolink Esthetic DC(VD), Panavia V5(PV). The same calibrated operator conducted the bonding procedures on moist dentin using self-etch mode according to the manufacturers' instructions. Bluephase G2 (Ivoclar, Amherst, NY) used for polymerization for 10s. A custom fabricated shear bond jig (Ultradent, South Jordan, UT, USA) was used for bonding procedures. Teflon mold was filled with the respective resin cement in a single increment, no greater than 2 mm. Shear bond strength was determined using a universal testing machine (Instron, Canton, MA, USA) at a cross-head speed of 1 mm/min with a load cell of 1,000 lbs, after 24-h storage (100% humidity-37°C) in the dark incubator. Specimens weren't submerged in water during this storage period. One-way ANOVA and Tukey test($p<0.05$) used to evaluate the effect of adhesive on shear bond strength and to identify these differences.

Results: Results are displayed in Table 1. There were no statistically significant differences found between the groups.

Resin Cement	Mean SBS (MPa)	Grouping
RelyX Universal	25.53	A
Calibra Ceram	25.10	A
Maxcem Elite	25.03	A
Variolink Esthetic	20.47	A
Panavia V5	17.89	A

Conclusion: OU360 is compatible with various dark-cured resin-cements in self-etch mode.

INFLAMMATORY EFFECT OF *PORPHYROMONAS GINGIVALIS* ON HUMAN PERIODONTAL MESENCHYMAL STEM CELL

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Objectives: Periodontitis is a widespread chronic inflammatory disease. Interestingly, bone regeneration rarely occurs even after successful periodontitis therapy, suggesting that periodontal Mesenchymal Stem Cells (MSCs) regenerative potential is deteriorated in periodontitis. *Porphyromonas gingivalis* (Pg), a keystone pathogen of periodontitis, promoting inflammatory bone loss. Pg expresses the peptidylarginine deiminase (PPAD) enzyme which can cause Histone citrullination and induction of inflammatory response. It is unclear if Pg can infect and affect the multipotency of local MSCs. The focus of this investigation was to study how *Pg* influences host stem cells on the cellular level and the role of monocytes and inflammatory cytokines.

Methods: Previously isolated human Periodontal-MSCs (HPD-MSCs) were cultured. To investigate, we established and analyzed a triple-culture system of MSCs exposed to Pg and co-cultured with THP1 monocyte cells. PPAD, anti-PPAD-mAb and glycyrrhizin (HMGB1 inhibitor) were additionally tested. The IL-1β level was tested by ELISA assay.

Results: Results showed that exposure of HPD-MSCs with Pg W83 induced atypical lytic cell death. An exposure of Pg W83 to HPD-MSCs and monocytes individually did not induce IL-1β production from HPD-MSCs. However, when tested MSCs exposed to Pg W83 and co-cultured with monocyte cells, significant elevated production of IL-1β was detected ($P<0.05$), while *Capnocytophaga gingivalis* tested as control did not promote a significant IL-1β production. Glycyrrhizin (HMGB1 inhibitor) and anti-PPAD mAb significantly inhibited the IL-1β production in HPD-MSCs exposed to Pg and co-cultured with monocyte cells.

Conclusion: In conclusion we hypothesized that Pg PPAD enzyme might be involved in atypical lytic cell death thus histone citrullination and chromatin release contribute to extracellular release of HMGB1 that can act on TLRs expressed on monocytes and induce the proinflammatory cytokine IL-1β. In conclusion Pg can modulate monocyte proinflammatory cytokines via Lytic Infection of HPD-MSCs. The following results may lead to the elucidation of mechanism underlying retarded bone regeneration in periodontitis.

AN INTERPROFESSIONAL SCHOOL-BASED INITIATIVE TO INCREASE ACCESS TO ORAL HEALTH CARE IN UNDERSERVED FLORIDA COUNTIES.

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Objective: To examine and describe the effect of an oral health education program on school-based nurses' acquisition of oral health knowledge.

Methods: Three-hour synchronous videoconference sessions provided training for nurses to conduct oral health risk assessments, screen for oral diseases, deliver oral health education, apply fluoride varnish, and refer children identified in need of further assessment and treatment to a dentist. Oral health knowledge acquisition was assessed by comparing pre-training and post-training examination scores. Analyses included descriptive statistics and the Wilcoxon Signed-Rank Test.

Results: All seventeen licensed school-based nurses from three Florida rural counties that have been entirely designated as Dental Health Professional Shortage Areas by the Health Resources and Services Administration participated in the oral health education training program. Analyses of the school-based nurses' test results showed a significant increase in correct answers on the post- training test (93%) compared to the pre-training test (56%) ($p \leq 0.001$). Six hundred forty-one children from six elementary public schools received oral health education, oral screenings, and fluoride varnish applications. Fifty-eight percent of children had untreated caries, 43% had treated caries, 15% had sealant on permanent molars, and 3% required urgent care.

Conclusions: The synchronous videoconference oral health training program was effective in improving school-based nurses' oral health knowledge. The knowledge acquired by school-based nurses via oral health training programs can be leveraged to increase access to oral health care for vulnerable and unserved school-aged populations.

SUPPRESSIVE EFFECT OF INOSINE ON RANKL-MEDIATED PERIODONTAL BONE RESORPTION

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Objective: It is well-established that in extracellular ATP (eATP) degradation pathway, pro-inflammatory eATP is converted to anti-inflammatory extracellular adenosine (eADO) by ectoenzymes, CD39 and CD73. Although eADO is further degraded to Inosine (INO) by ectoenzyme adenosine deaminase (ADA, CD26) which is reported to be expressed in gingival tissue, the possible role of eINO in periodontitis is unknown. Importantly, INO is metabolically more stable (half-life 15h) than ADO (half-life <10 s). The purpose of this study is to evaluate the effect of eINO on osteoclast-mediated bone resorption in periodontitis in comparison to that of ATP and chemical mimetic of ADO, 5′-N-Ethylcarboxamido Adenosine (NECA).

Methods: Murine RAW264.7 macrophage-like cells were stimulated *in vitro* with RANKL (10 ng/ml) with or without ATP, INO or NECA (4-days for TRAP-staining, 7-days for pit formation assay). The possible cytotoxicity of respective reagent was examined in a dose response manner, and the range of concentration that doesn't affect the viability of RAW264.7 cells was determined. To evaluate the effect of respective reagent on osteoclastogenesis, TRAP staining and pit formation assays were performed. Statistical analysis was performed by Tukey post-hoc tests with ANOVA.

Results: The concentration up to 200 uM for INO and NECA, respectively, was found to be not toxic for RAW264.7 cells. Both INO and NECA significantly suppressed RANKL-induced osteoclastogenesis and pit formation ($P<0.05$), whereas ATP promoted RANKL-induced osteoclastogenesis and pit formation. However, within the range of concentrations assessed (100 uM- 200 uM), the level of osteoclastogenesis suppressions was significantly higher by NECA than INO ($P<0.05$). Finally, the ATP-mediated upregulation of osteoclastogenesis was suppressed by NECA as well as INO.

Conclusion: The present study demonstrated that INO can suppress RANKL-induced osteoclastogenesis and bone resorption, and that INO can counteract the ATP-mediated upregulation of osteoclastogenesis, suggesting that INO can be a novel therapeutic target for periodontitis.

ACTIVATION OF PIEZO1 IN OSTEOCLASTS INHIBITS BONE RESORPTION IN PERIODONTITIS

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Objectives: Irreversible inflammatory bone resorption is characteristic to periodontitis. An emerging body of evidence supports that mechano-sensitive Ca^{2+} ion channel Piezo1 plays important regulatory roles in a variety of cellular activities. However, the role of Piezo1 in osteoclast (OC)-mediated bone remodeling in periodontitis remains elusive. The aim of this study is to investigate the effect of Piezo1-elicited mechano-sensory transduction on OC-mediated bone remodeling in periodontitis.

Methods: Murine bone marrow-derived mononuclear cells were cultured with or without M-CSF (25 ng/ml), RANKL (10 ng/ml), Yoda1 (Piezo1 activator, 5 µM) and/or shear flow generated by an orbital shaker (30 rpm). OC-genesis and related gene expressions were determined by TRAP staining and pit formation assay, as well as qPCR. MC3T3-E1 osteoblast (OB)-progenitor cells were exposed *in vitro* to the supernatant of Yoda1-treated OCs with or without anti-IGF-1-mAb. OB-genesis-mediated mineralization was evaluated by alkaline phosphatase (ALP) activity and Alizarin red staining. Yoda1 was injected systemically to the mice with ligature induced-periodontitis. Then, bone resorption and OC-genesis related gene expressions were monitored at Day-7.

Results: The development of multi-nucleated TRAP-positive cells and pit area were decreased by treatment of BMMCs with Yoda1 or shear flow. OC-genesis related genes including *Ocstamp, Acp5 and Mmp9* were downregulated by Yoda1 (P<0.01), while *Igf1* gene was upregulated (P<0.01). Mab-based neutralization of IGF-1 in the supernatant of Yoda1-treated OCs reduced both ALP activity and mineralization in MC3T3-E1 cells. In the mouse model of periodontitis, Yoda1 administration significantly suppressed bone resorption (P<0.05) and OC-genesis related gene expressions (P<0.05), while *Igf1* gene expression was upregulated (P<0.05).

Conclusions: The present study demonstrated that the activation of Piezo1 can decrease OC-genesis, while promoting the production of coupling factor, IGF-1 from OC precursors. Furthermore, Yoda1 abolished bone resorption in the murine periodontitis model. These results suggest that Piezo1 can be a molecular target for periodontitis therapy.

ANATOMICAL CONSIDERATIONS IN ENDODONTIC SURGERY OF THE PALATAL ROOT OF MAXILLARY MOLARS: A REVIEW

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Introduction: The goal of endodontic treatment is to prevent, intercept and treat pulpal/periapical pathosis. When non-surgical treatment is not possible, the clinician should consider an apicoectomy as part of endodontic retreatment regimens. One of the most challenging conditions in endodontic surgery is management of apicoectomies in the posterior region, such as the palatal root of maxillary molars. Clinicians are often unwilling to use the palatal approach in apical surgery for the maxillary first and second molars (Lee et al., 2020).

Objectives: Determine the anatomical structures when performing endodontic surgery in the palatal root of maxillary molars. This traditional literature review guides clinicians to perform the maxillary molars palatal root approach in a predictable way in endodontic surgery.

Methods and Materials: Using data base as Pubmed, Google Scholar and Web of Science search was done using the key words and Boolean operators (AND, OR) to conduct a literature review about maxillary posterior endodontic surgery.

Results: Apical surgery on the palatal root of the maxillary molar using a palatal approach can be predictable and successful, complications nowadays can be minimal.

Conclusion: The palatal and the transantral approach offer viable options for performing an apicoectomy on the palatal root of maxillary molars. The decision on the approach should be based on a comprehensive assessment of the individual case, considering anatomical factors, the surgeon's expertise, and patient-specific considerations.

INFLUENCE OF MATERIALS & METHOD OF CONVERSION ON INTERIM PROSTHESIS FRACTURE STRENGTH

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Objectives: To demonstrate that denture base material and method of conversion technique are significant factors that determine the functional fracture strength (durability) of the immediate load prosthesis.

Methods: Thirty samples of each material were used per conversion technique. Thirty Ivoclar Acrylic injection molded blocks for conventional conversion and thirty Ivoclar Acrylic injection molded blocks for Smart Denture Conversion (SDC) protocol. Thirty Avadent milled blocks for conventional conversion and thirty Avadent milled blocks for Smart Denture Conversion (SDC) protocol. Thirty SprintRay OnX printed resin blocks for conventional conversion and thirty SprintRay OnX printed resin blocks for Smart Denture Conversion (SDC) protocol. All samples were picked up with UNIFAST TRAD polymer & monomer during the denture conversion. Then gradually finished with a 600 grit size sandpaper. Fracture strength of each specimen will be tested. Each specimen will be taken to failure using a 3-point bend test on an Instron machine with a static load test (Instron, Canton, MA, USA). A two-way ANOVA with Material (3D Printed Resin, Injection PMMA, Milled PMMA (Avadent), Method of Conversion (Conventional, Smart Denture Conversion), and the interaction of Material by Method of Conversion was conducted with a false-discovery post-hoc comparisons. Controlling for the false discovery rate (FDR) is a way to identify as many significant features as possible while incurring a relatively low proportion of false positives. JMP 16.0 was used in the analysis and a p value less than 0.05 are considered statistically different.

Results: Results from this study are described as: method of conversion, material, and method of conversion*material. Method of conversion was not statistically significant for mean fracture strength. Resulting in a P value < 0.145. Choice of material showed statistically significant results for mean fracture strength with a P value < 0.001. Combination of Method of Conversion * Material showed a statistically significant result for mean fracture strength with a P value < 0.001. Summary statistics are presented in Table One. The ANOVA table with omega-squared (effect size) are presented in Table Two. Pairwise comparisons are presented in Table three. A significant difference was found by material and the interaction of method of conversion by material. Reviewing the Omega squared indicates that the interaction of Method of Conversion by Material accounts for 11% of the variation, Material 6%, and Method of Conversion (not significantly different) 1%.

Conclusion: In conclusion, the samples made from 3D printed resin converted using the "Smart Denture Conversion" technique had the mean highest fracture strength. The lowest mean fracture strength recorded were samples made from Ivoclar Injection acrylic converted using the conventional conversion technique. The mutilation of denture base through required "through and through" access holes during the conventional conversion technique is a contributing factor

towards mean fracture strength. Based on this study, the author advises dental clinicians to adopt the "Smart Denture Conversion" technique in combination with 3D printed resin for fabrication of reliable and strong interim conversion prosthesis.

*Tables & Figures of Data & statistics are on following page. If 1 page limit for abstract, do not include pg. 2
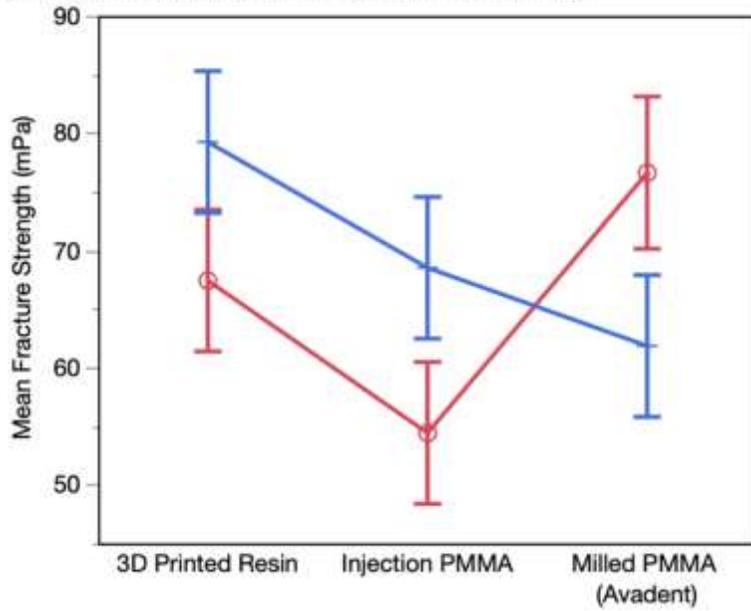
Table 1. Summary Statistics

		Conventional	Smart Denture Conversion
3D Printed Resin	N	30	30
	Mean	67.41	79.23
	SD	16.88	20.99
	Min	44.28	42.56
	Max	98.51	120.68
Injection PMMA	N	30	30
	Mean	54.40	68.50
	SD	14.40	18.12
	Min	19.91	31.57
	Max	80.97	99.52
Milled PMMA (Avadent)	N	26	30
	Mean	76.62	61.83
	SD	10.13	17.38
	Min	59.19	19.40
	Max	99.94	95.59

Table 2. ANOVA and Effect Size Table

Source	DF	Sum of Squares	F Ratio	Prob > F	Omega Squared
Method of Conversion	1	605.17	2.15	0.145	0.01
Material	2	4353.92	7.73	0.001	0.06
Method of Conversion*Material	2	7360.52	13.06	<.0001	0.11

Figure 1. Mean Comparisons with 95% Confidence Intervals

ANTIBIOTICS PRESCRIBED BY MEMBERS OF THE AMERICAN ASSOCIATION OF ENDODONTICS: A 2023 REPORT OF A NATIONAL SURVEY
Steffi Estevez, DDS

Introduction: The purpose of this study was to assess the antibiotic prescribing patterns among active members of the American Association of Endodontics (AAE).

Methods: A 15-question survey was sent via https://redcap.nova.edu to 3879 active members of the AAE. The survey was designed to address the new 2020 AAE guidelines for the prescription of antibiotics in situations where patients claim to be allergic to penicillin. The data was analyzed using descriptive statistics and chi-square analysis.

Results: The survey received a response from 418 endodontists, indicating a response rate of 10.10%. On average, 25.4 patients were treated per week. Amoxicillin was the most frequently prescribed (86%) followed by clindamycin (66.1%). For patients who reported a penicillin allergy, 72.4% of responded yes for Clindamycin and 68.6% stated they would prescribe Azithromycin (Z-Pak). Respondents reported prescribing antibiotics for irreversible pulpitis with moderate symptoms (0.8%), necrotic pulp/symptomatic apical periodontitis with moderate to severe pre-op symptoms (17.8%%), chronic apical abscess without (5.6%) or with symptoms (14.9%), acute apical abscess (54.3%), incision and drainage with diffuse intraoral and extraoral swelling (24.7%), avulsion (19.3%), endodontic surgery (11.9%), retreatment (4.9%), and postoperative pain after instrumentation or obturation (4.8%)

Conclusions: Since 1999, there has been a notable transition in the preferences of endodontists regarding antibiotics. There has been a substantial shift from prescribing penicillin V to favoring amoxicillin as the primary choice of antibiotic. Additionally, there has been an increase in the utilization of Azithromycin instead of clindamycin for patients with a penicillin allergy.

OCCLUSAL VARIABLES PRESENT IN THE NORMAL POPULATION COMPARED TO PEOPLE WITH TEMPOROMANDIBULAR DISORDER
Gertler, A., Menchel, H.

Objectives: To compare occlusal determinants present in the normal population to two hundred patients with Temporomandibular Disorders who were seen in a Facial Pain Clinic.

Methods and Materials: Occlusal determinants documented in NSU CDM Facial Pain Clinic of patients presenting with TMD were compared to the normal population. The norms were determined from literature review of peer reviewed articles.
Inclusion criteria included patients presenting to the clinic complaining of TMD symptoms and who were examined according to the revised (2014) Diagnostic Criteria for TMD (DC/TMD). Only patients with joint pain according to type II and II DC/TMD criteria were included.
Exclusion criteria were people with only muscle symptoms, history of trauma, people who had recent orthodontic treatment or recent full mouth rehabilitation. Only patients with complete dental arches were included. The data comprises several parameters of occlusion for each patient including (i) angle classification on the right and left side, (ii) presence of CR-MICP slide, (iii) the excursive guidance, (iv) bruxism status and (v) TMD diagnosis. Anterior determinants including overbite and overjet and midline. In comparing these patients to each other and to the normal population, we will determine if any associations can be made which can support or refute previous studies.

Results: Occlusal factors amongst the population of patients with TMD, compared to accepted norms, were different and statistically significant. Within the entire sample set, there was a statistically significant incidence of bruxism. 162 of the 200 patients presented with bruxism. Compared to the norm, this yielded a Chi square of 465 with 1 degree of freedom, or a $p < 0.0001$. Within the population of patients presenting with Osteoarthritis, 85.9% of them were bruxers, whereas only 60% of the patients with disk displacement with our reduction were bruxers. Within the entire sample set, there was a statistically significant disparity of angle classification. 70% of the sample was class I, 18.5% was class II, and 11.5% was class III. Angle classification compared to the norm yielded a Chi square of 29.5 with 2 degrees of freedom, or a $p < 0.0001$. When comparing overjet and overbite, there was no statistically significant difference. All categories were within one standard deviation of the mean. Disk replacement without reduction showed a much higher percentage of patients who were class III at 20%, and only 66.7% were class I. Only 9.9% of the the patients with arthralgia were angle class III, and 72.5% of them were angle class I. There was no statistically significant difference among patients with a CR-MICP slide. There was a difference between the average age of each diagnosis. The osteoarthritis group had an average age of 59.47, the arthralgia patients was 48.35. Meanwhile, the average age of the disk displacement with reduction patients was 36.45 and the average age of disk displacement without reduction was 32.33.

Conclusion. A statistically significant relationship between angle classification, age, and bruxism and TMD patents, is present. However, a causal relationship should not be overstated; differences in occlusal factors may be a consequence of rather than a cause for TMD. There was no statistically significant difference between overjet, overbite, or CR-MICP slide.

INVOLVEMENT OF PIEZO1 MECHANOSENSITIVE RECEPTOR IN CELLULAR SENESCENCE IN PERIODONTITIS.

Ahmadreza Khademi, Satoru Shindo, Shin Nakamura, Tomoki Kumagai, Alireza Heidari, Maria Rita Pastore, Elizabeth Leon, Maria Hernandez, Saynur Vardar-Sengul, Toshihisa Kawai

Objectives: Aging exacerbates pathogenic outcomes and progression of periodontitis. It is true that aging also occurs at the single-cell level (cellular senescence) which can be induced by bacterial component, such as LPS. Although emerging body of evidence supports that mechanosensitive Ca^{2+} ion channel, Piezo1, plays a relevant role in regulation of a variety of cellular events, the possible engagement of Piezo1 in periodontitis is largely unknown. The aim of this study is to establish the role of Piezo1 in *Porphyromonas gingivalis* (*Pg*)-derived outer membrane vesicles (*Pg*-OMVs)-induced cellular senescence.

Methods: *Pg*-OMVs were isolated from *Pg* W83 using an ultra-centrifuge (100,000xg, 1 hour). Human gingival fibroblasts (HGFs) isolated from healthy donors were stimulated with or without *Pg*-OMVs (1 ug/ml) and/orYoda1 (Piezo1 activator, 5 uM) under the shear flow (rotating shaker, 15 degrees, 30 rpm) or static condition. To evaluate the level of cellular senescence induced in HGFs, β-galactosidase staining was performed. Productions of Senescence-Associated Secretory-Phenotype (SASP) factors, Interleukin (IL)-6 and IL-8, were measured using ELISA and q-PCR. Statistical analysis was performed by one-way ANOVA as well as Tukey-Kramer's multiple comparison tests.

Results: Exposure of HGFs to Yoda1 and mechanical stress generated by shear flow increased the expression of protein and mRNA for IL-6 and IL-8 ($P<0.05$) as well as that of β-galactosidase ($P<0.05$). Yoda1 showed a strong additive effect on Pg-OMVs-induced productions of proteins and mRNA for IL-6 and IL-8 as well as β-galactosidase expression compared to *Pg*-OMVs or Yoda1 alone ($P<0.05$). Very importantly, the effect of YODA1 on all those outcomes were proportional to those caused by shear flow, suggesting that Piezo1 is engaged in the sensing of mechano-stress by HGFs.

Conclusions: The present study demonstrated that piezo1-mediated mechano-stress-sensing can upregulate the *Pg*-OMVs-induced cellular senescence in HGFs suggesting that mechanical stress in periodontitis may accelerates cellular aging and inflammatory tissue destruction.

BUILDING RESILIENCE IN DENTAL STUDENTS: A NOVEL BIOPSYCHOSOCIAL CURRICULUM

Siegel, S. C., Kandalam, U., Newman, R., Gannavaram, S., Cummings, N.

Objectives: This pilot study investigated the feasibility and effectiveness of the SKY Breath Happiness Program (SBHP), a bio-psycho-social curriculum for promoting stress-management, resilience building and well-being in first year dental students (D1s). The SBHP emphasizes a controlled breathing sequence called Sudarshan Kriya Yoga (SKY), documented to increase parasympathetic tone, and to offer faster recovery ("resilience") of corto-limbic networks after affective stimulation.

Methods: An NSU IRB was granted with exempt status (2021-334-NSU). SBHP was introduced via emails before the D1 first semester. Recruiting involved a presentation during student orientation explaining SBHP was to be conducted over three 3-hour sessions of week-one classes, with a reinforcement class one week later. Informed consent was obtained from enrolled participants (N=15; intervention group). Non-enrolled D1s (N=103;control group) read a waived consent (not of process). This mixed-methods study included an online anonymous survey using REDcap both pre-course and mid-semester (eight weeks post course). Assessment for enrolled students included themes extracted from journaling completed one week post-intervention.

Enrolled and non-enrolled D1 students completed four standardized measures; 1). Perceived Stress Scale; 2). PANAS Positive and Negative Affect Scale; 3). Social Connectedness Scale; 4). Connor-Davidson Resilience Scale, pre-course and eight weeks post-course (i. e. after mid-term exams). Pre- and post course results are measured and evaluated using descriptive statistics for study variables. Differences between the groups were analyzed using a student's t-test. An a priori value of ≤ 0.05 was used for significance. Intervention student experiences were independently scored for themes by two investigators.

Results: Fifteen students enrollees and 103 non-enrollees completed the pre-course surveys (91% participation). The post-course survey response rate was 56%. 100% of enrolled students completed the open-ended journal. Themes extracted from journaling showed aspects of global well-being (Figure 1) and long term anticipated resilience. Further, 100% of the enrollee respondents rated the course as very good or excellent in the standard University course evaluation. Quantitative data are undergoing more complete analysis. Only qualitative/descriptive data are reported.

Conclusion: There was a 91% class participation in the pre-course survey and a 56% participation post-course surveys. A higher percentage of minority students enrolled in SBHP than whites or non-enrollees. Students rated the course as very good (highest rating) and as contributing to their overall mental well-being.
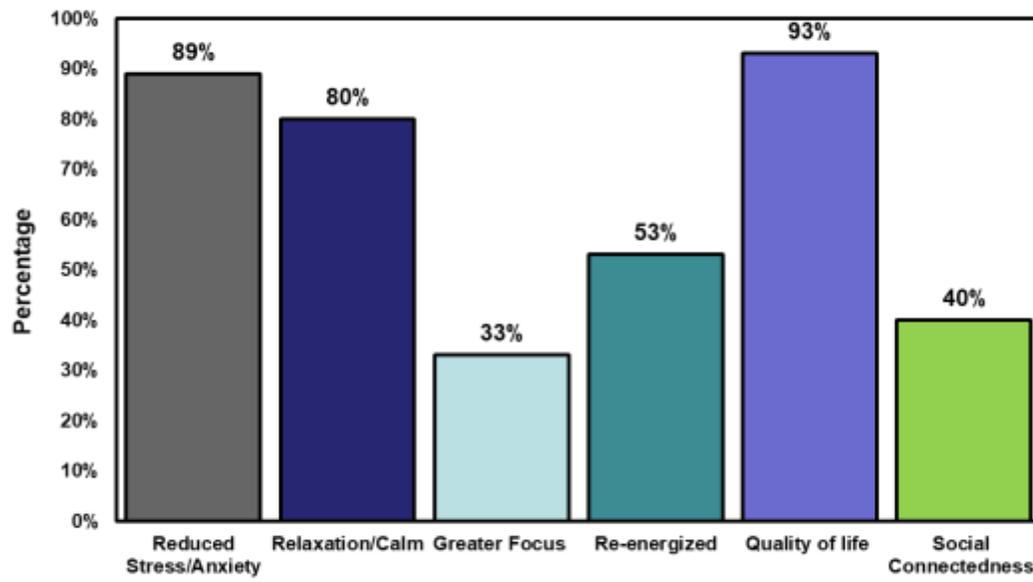
Figure 1.

A REVIEW OF CALCIUM HYDROXIDE AND OTHER INTRACANAL MEDICAMENTS IN SINGLE VERSUS MULTIVISIT ROOT CANAL TREATMENT

Yousof Sinjab DMD, Daniel Mizrahi BS, Yehuda J. Benjamin DMD, FAGD, MS

Objective: The purpose of this review is to review the mechanism of actions of intracanal medicaments (Calcium hydroxide, Chlorhexidine, Triple antibiotic) and compare the success rates when used in endodontic root canal therapy. This will provide an overview and help familiarize the readers with these intracanal materials, understand which intracanal materials have a superiority in intracanal medicaments, and help increase success when used in endodontic treatments by educating the general dentists and specialist.

Materials and methods: Databases of Journal of Endodontics and PubMed were searched to conduct a literature review on intracanal medicaments in single vs multivisit treatment.

Results: All three intracanal medicaments have successful outcome on eliminating intracanal bacteria. Calcium hydroxide (CaOH) has proven to inactivate endotoxins however its effect on *enterococcus faecalis* and biofilms continues to be a controversy. 2% Chlorhexidine (CHX) and chlorhexidine gel has a higher success rate removing gram-positive bacteria (specifically *enterococcus faecalis)* than calcium hydroxide. Triple antibiotic (TAP) possess a negative side effect on stem cells of the apical papilla (SCAP) and dentin staining.

Conclusion: The use of intracanal medicaments CaOH, CHX, and TAP have shown to be effective in endodontic treatments. The use of CaH and its efficacy remains questionable. Some studies suggested the use of CHX as alternative instead of CaH. Chlorhexidine in the form of a gel or 2% solution alone (with no additional additives such as CaOH) have a higher success rate in removing e*nterococcus faecalis* than CaOH. TAP showed to be effective in eradicating endodontic microbes. However, the negative side effects on SCAPs and dentin staining makes other alternative intracanal medicaments favorable. Further research is required to clarify intracanal medicaments antimicrobial mechanisms effectiveness in endodontic treatments.

ODONTOGENIC MYXOMA: REPORT OF A CASE
Chiang, E., Solomon, L., Morón, E.

Introduction: Clinical photos, panoramic radiographs, Cone Beam CT imaging and photomicrographs are used to illustrate the case of a maxillary odontogenic myxoma that was diagnosed and treated at Nova Southeastern University College of Dental Medicine. Odontogenic myxoma is a benign tumor with a reported incidence of 0.07 / per million population. It represents only 3-6% of odontogenic tumors.

Case Presentation: A 32-year-old African-American woman presented to the NSU CDM oral surgery clinic for follow-up of a lesion of one-year duration. She said the swelling on her palate was previously biopsied at an outside provider, and she was told that it was "benign". Initial intraoral clinical examination showed a large erythematous swelling on the left side of hard palate which extended to the soft palate and slightly crossed the midline to the right. The lesion was tender to palpation, indurated, and non-ulcerated. The panoramic radiograph showed a unilocular radiolucent lesion in the posterior left maxillary region. The CT scan demonstrated a large expansile/erosive mass involving the left maxilla and soft tissues left side of the face with expansion into the floor of the left maxillary sinus. The lesion measured 4.4 cm transverse by 3.8 cm craniocaudal by 4.2 cm AP. An incisional biopsy with histologic examination revealed an infiltrative proliferation of paucicellular myxoid tissue composed of scattered spindled fibroblasts with tapered nuclei among scattered collagen fibers and abundant vacuolated basophilic stroma. The histologic diagnosis was odontogenic myxoma. Treatment consisted of left maxillary resection with reconstruction of left infrastructure maxillectomy/palate/alveolar arch defect with a free left osteocutaneous fibular flap, fixation of fibula flap to maxillary alveolar arch and zygoma, split-thickness skin grafts (STSG) from left thigh to left leg fibula donor site, and vacuum assisted closure (VAC). All surgical procedures were performed under general anesthesia. After surgery, the patient was in ICU for close monitoring and flap checks. A dietitian assessed nutritional needs and a speech and language therapist assessed the patient to suggest exercises to help with patient speech. The patient was transferred to the floor, and discharged from hospital eight days later. A comprehensive postoperative clinical examination including a postoperative panoramic radiograph revealed satisfactory results from the surgery. Unfortunately, the patient was lost to further follow-up.

Conclusions: The head and neck area has a complicated anatomy with many vital structures in close proximity to each other. Maxillary tumors may grow into the adjacent sinus and greatly enlarge before causing clinical signs and symptoms. Surgical removal of large maxillary tumors is difficult and the reconstruction is complex. This case highlights the importance of communication, so the patient understands their diagnosis and the next steps for treatment. The "benign" diagnosis this patient was given by an outside provider did not mean that a delay in treatment is acceptable. By the time she was evaluated in the oral surgery clinic at Nova Southeastern University College of Dental Medicine, her disease was at an advanced stage. If it had been

appropriately treated when she first presented to an outside provider a year earlier, she may have been spared such an extensive surgery, reconstruction and rehabilitation.

No conflicts of interest are reported by the authors and all patient identifiers are erased.

PORPHYROMONAS GINGIVALIS OUTER MEMBRANE VESICLES EXACERBATE RETINAL MICROVASCULAR ENDOTHELIAL CELL DYSFUNCTION IN DIABETIC RETINOPATHY

Shengyuan Huang, Sunniva Ruiz, Satoru Shindo, Shin Nakamura, Toshihisa Kawai, Xiaozhe Han.

Objectives: Diabetic retinopathy (DR) is manifested by a series of pathological changes in retina including the altered permeability of the blood-retinal barrier (BRB). Current epidemiological studies provide limited evidence suggesting that periodontitis is associated with DR. However, there is a lack of basic research elucidating how periodontitis affects the severity of DR. This study is to determine the effect of *Porphyromonas gingivalis (P.gingivalis) outer membrane vesicles* (OMVs) on the BRB integrity in diabetic retinopathy.

Methods: C57BL6/J mice (6 weeks, male) were divided into four groups: Control, DR, DR+OMVs, and OMVs (n=8), and were injected with STZ (120mg/kg) for DR model. After 12 weeks, *P.gingivalis* (ATCC33277) OMVs (100μg) was injected into mice through tail vein twice a week for 8 weeks. The total retinal microvascular area and number of microaneurysms were evaluated by fundus fluorescein angiography (FFA). Retina microvessel diameter and pericyte number were assessed by retinal microvascular staining. Cultured human retinal microvascular epithelial cells (hRMECs) were stimulated with *P.gingivalis* OMVs (50μg/ml) for 24h. Cell permeability was evaluated by fluorescein dextran assay. Cell apoptosis was analyzed by Ratiometric Membrane Asymmetry Probe and Mitochondrial Membrane Potential Probe. RT-qPCR was used to detect the expression of cell permeability, apoptosis, and inflammation-related genes. In addition, protease-activated receptor-2 (PAR-2) regulated OMVs-induced TNF-α, MMP-9 mRNA expression, cell death, and endothelial permeability.

Results: The total microvascular area and the number of microaneurysms in the DR+OMVs group were significantly increased compared to the DR group, with no significant difference between the OMVs group and the control group. The diameter of the microvessel and the number of pericytes in the DR+OMVs group were significantly lower than in the DR group. The OMVs-treated group demonstrated an increased leakage of fluorescently-labeled dextran through cultured hRMECs, and an increased percentage of apoptotic cells compared to the control group. Expressions of ICAM-1, IL-6, MMP-9, TNF-α, IL-1β, and MMP-9 were significantly increased in the OMVs-treated group than in the control group. There was no difference in the expression of MMP-2, ve-cadherin, and TGF-β. *P. gingivalis* OMVs induced proinflammatory cytokine production, mitochondria-related cell death of HRMECs and the endothelial dysfunction were diminished in the presence of PAR-2 inhibitor AZ3451.

Conclusions: This study is the first research report to delineate that *P. gingivalis* OMVs may compromise retina microvascular integrity via increased inflammation and endothelial dysfunction, hence exacerbating DR pathogenesis. It provides a model system for expanded research in the field of microbial OMVs as a link between oral and systemic diseases.

P. gingivalis OMVs increase number of retinal microaneurysms in DR mice
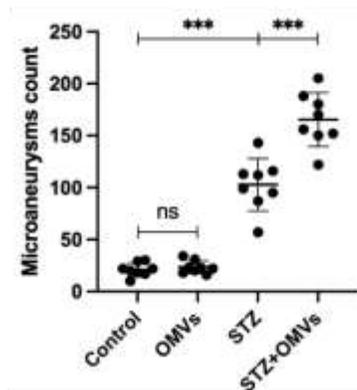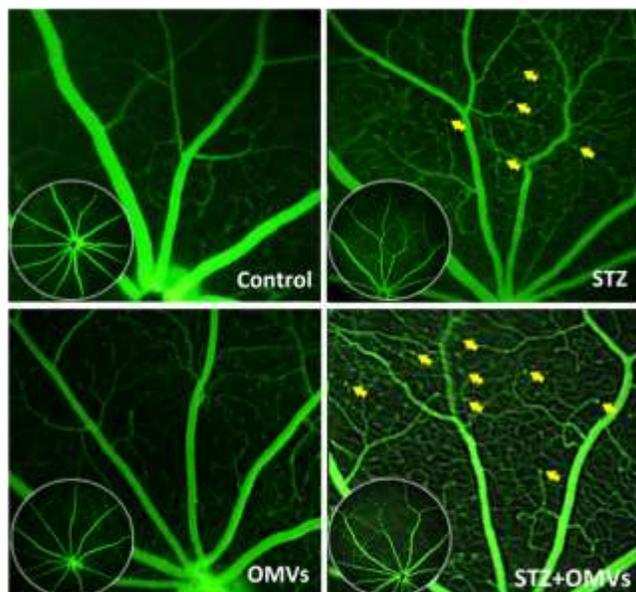
FFA showed an increased number of microaneurysms in the STZ+OMVs group than in the STZ only group

MEASURING THE DISTANCE FROM THE CROWN MARGIN TO THE ALVEOLAR CREST BONE FROM CLINICAL INTERPROXIMAL RADIOGRAPHIC IMAGES: A PILOT STUDY

Mesko, M., Dobrin, R., Vazquez, J., Garcia, M., Orozco, J., Caldas, B., Walia R, Youseff, Y., Elderbashy, I, and Mizrahi, S.

Objectives: To assess the level of agreement of different evaluators when measuring the distance from the crown margin/cement-enamel-junction (CEJ) to the alveolar crest bone (ACB) from clinical radiographs.

Significance: The evaluation of clinical radiographs should be highly agreeable amongst all clinicians, and proper methods of radiographic evaluation and diagnosis are required for research or educational purposes.

Methods: As an inclusion criterion the radiographs had to show a tooth that showed a prosthetic crown surrounded by both adjacent teeth (restored or not). In this study, 10 de-identified bitewing radiographic images were analyzed by 6 different operators (two periodontists, two team leaders, and two D4 predoc students). The observers were instructed and calibrated to use the *XDR ruler tool* (in millimeters) for the following measurements/variables: (1) the distance between the CEJ to the ACB on the distal surface of the mesial adjacent tooth; (2) distance between finish line of a crowned tooth to its ACB on the mesial and (3) distal surfaces and (4) the distance between the CEJ to the ACB on the mesial surface of the distal adjacent tooth (Figure 1). The evaluators, who were not blind to the objective of the study, independently measured the set of images within 24 hours after the meeting. A after a two-week washout period, the same set was measured again. The data was recorded and descriptive statistics such as mean, standard deviation, median were calculated, and percentiles were used to summarize the data. For the inter-observer agreement, the Friedman Test and Kendall's W Concordance Coefficient were used. Statistical measurements were performed using SPSS, and the significance level was 5% (2-tailed).

Results: An excellent correlation or agreement between the evaluators when evaluating the four measurements ($p < .05$) was found, except on measurement 3, which nonetheless still showed a good correlation in both statistical tests performed.

Conclusion: The level of agreement between the evaluators was excellent, showing that they are calibrated. The authors suggest a future study involving a higher number of radiographic images to determine possible sources of bias.

Figure 1. Cropped bitewing radiograph with the respective variables and measurements (in mm).
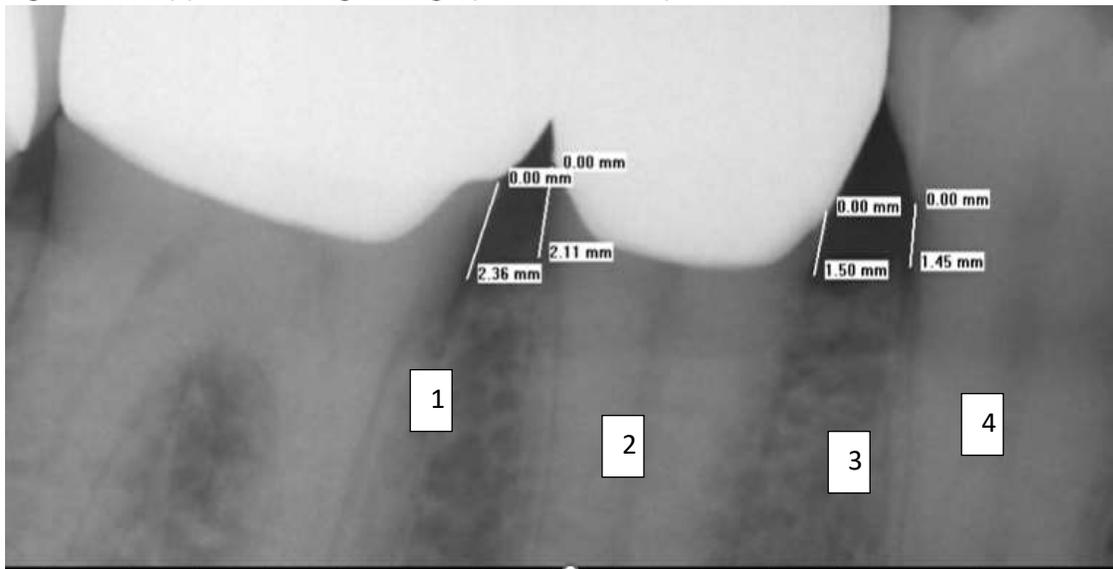
	Variable	Variable	Variable	Variable
Case	1	2	3	4
80G	2.36	2.11	1.5	1.45

Table 1. Descriptive analysis. Summarization of the data for the different evaluators.

	N	Mean	Std. Deviation	Minimum	Maximum	Percentiles		
						25th	50th (Median)	75th
Evaluator1	10	1.8550	.86920	.43	3.27	1.1075	1.9350	2.5250
Evaluator2	10	1.9090	.80816	.63	3.27	1.4225	1.7900	2.4850
Evaluator3	10	1.5340	.90992	.23	3.33	.7500	1.4900	2.0475
Evaluator4	10	1.6730	.84074	.22	2.92	1.2525	1.5450	2.4675
Evaluator5	10	1.7910	1.03570	.41	3.25	.7300	1.7000	2.8900
Evaluator6	10	1.7850	.75949	.77	2.82	.9475	1.8050	2.4625

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