

26th ANNUAL

RESEARCH DAY



INDIANA UNIVERSITY SCHOOL OF DENTISTRY
April 9, 2018





Congratulations! Delta Dental would like to congratulate the Indiana University School of Dentistry students on their Research Day presentations.

Your hard work and dedication to dentistry are an inspiration to us all.

IUSD RESEARCH DAY PROCEEDINGS

VOLUME 26

APRIL 9, 2018

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This piece is a collage inspired by the link between science and art, which is research. Both fields require specific forms of creativity and critical investigation to successfully arrive at the moment of discovery. Often, as a by-product of the scientific process, beautiful imagery is created. However, it is frequently overlooked as it is the means to a larger exploration. This piece was created as a way to stop and honor that beauty, creativity, and skill found in these works. The perfect example of this link comes in the form of Leonardo da Vinci's Vitruvian Man, created in 1490. It is an illustration of the ideal human proportions and a representation of the golden ratio which is ubiquitous throughout nature and art. Da Vinci, an Italian Renaissance polymath, was not only a renowned painter but a scientist in his own right. He has had a lasting impact on a vast array of fields outside the fine arts, most specifically anatomy and physiology. As scientists, doctors, and creative thinkers it is easy to appreciate Da Vinci's remarkable foresight. This image is in honor of his love of both art and scientific study and the distance we have come since his time. It is composed of SEM and slide images of materials, bacteria, viruses, and tissues at varying scales so that one may appreciate the images for what they are other than evidence of a scientific process -- art.

Cover design and layout by Nicole Alderson.
Student Research Group photo and back cover by Abby Morgan.
Research Day proceedings monograph prepared by Keli Seering.

ABOUT OUR KEYNOTE SPEAKER

DR. RAUL I. GARCIA



Raul I. Garcia, D.M.D., is Professor and Chair, Department of Health Policy and Health Services Research at Boston University Henry M. Goldman School of Dental Medicine. He is also the director of the Center for Research to Evaluate and Eliminate Dental Disparities (The CREEDD), established at BU in 2001 and supported by the National Institutes of Health. The Center's aims are to identify the determinants of oral health disparities and to design and implement community-based interventions to eliminate oral health disparities. In addition, he is the Director of the U.S. Dept. of Veterans Affairs (VA) Dental Longitudinal Study (DLS), a decades-long investigation on aging and oral health. The VA-DLS has also examined

the role that oral conditions may play as risk factors for cardiovascular disease and other systemic health outcomes, and how oral conditions may affect health-related quality of life.

Dr. Garcia is a 1981 graduate of the Harvard School of Dental Medicine, where he received the D.M.D. degree summa cum laude. He completed his specialty training in Periodontology in 1984, and went on to receive a Master of Medical Sciences degree from Harvard in 1985.

In March 2018, he will complete his term as president of the American Association for Dental Research and then serve as the AADR Immediate Past-President until June 2019.

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Schedule of Events

IUPUI Campus Center 4th Floor

Thursday, April 5

5:00 pm – 8:30 pm Poster Judging (Dental School)

Monday, April 9

9:00 am Commercial Exhibition opens
(CE 450C)

9:00 am – 12:00 pm Continuing Education Event
Integrating Contemporary Digital Technology
into Your Practice
(CE 450A-B) **Dr. Dean Morton & Colleagues**
Department of Prosthodontics,
IU School of Dentistry

12:00 pm Registration Open (4th floor lobby)

1:00 pm Welcome Remarks
(CE 450A-B) **Dr. John N. Williams**
Dean, IU School of Dentistry

1:05 pm Opening Remarks **Dr. Simon Atkinson**
IUPUI Vice Chancellor for Research,
IUPUI School of Science

1:15 pm Introduction of Keynote Speaker **Dr. Tien-Min Gabriel Chu**
Associate Dean for Research,
IU School of Dentistry

1:20 pm Keynote Address
“Science and Dental Practice” **Prof. Raul Garcia**
Professor and Chair,
Boston University
School of Dental Medicine

1:50 pm IU School of Dentistry Research Update **Dr. Tien-Min Gabriel Chu**
Associate Dean for Research,
IU School of Dentistry

2:00 pm Announcement of Faculty Awards **Dr. John N. Williams**
Dean, IU School of Dentistry

2:10 pm Announcement of Poster Awards &
Presentation by Awardees **Dr. Frank Lippert**
President, Indiana Section, AADR

3:00 – 4:30 pm Research Presentations (CE 405, 406, 409)
3:00 pm - 3:45 pm: Odd-numbered Posters and Clinical Case Reports
3:45 pm - 4:30 pm: Even-numbered Posters and Clinical Case Reports

4:30 pm Announcement of Exhibitor Passport Winners
Removal of Posters
Commercial Exhibition closes



IUPUI

SCHOOL OF DENTISTRY

OFFICE OF THE DEAN

Indiana University–Purdue University
Indianapolis

April 9, 2018

Indiana University School of Dentistry Colleagues and Friends:

Welcome to the 26th Annual Indiana University School of Dentistry Research Day.

IUSD has a long history and proud tradition of creating new knowledge through research and discovery to improve oral health care for all. I am proud of our students and their faculty mentors for presenting their scientific endeavors today. Our annual Research Day showcases their accomplishments and allows us to look beyond a single research focus to the wider horizon of translating scientific discoveries into improved oral health care.

We are most pleased to welcome Dr. Simon Atkinson, IUPUI vice chancellor for research and Chancellor's Professor in the School of Science, to deliver opening remarks. We are also honored to have with us today as our keynote speaker Dr. Raul Garcia, professor and chair of the Department of Health Policy & Health Services at the Henry M. Goldman School of Dental Medicine at Boston University. Dr. Garcia is research director of the Northeast Center for Research to Evaluate and Eliminate Dental Disparities. His career path demonstrates the spectrum of options open to dental researchers. Dr. Garcia's initial focus was periodontal disease, then he transitioned into public health research and policy. I know you will enjoy his message, and I encourage you to attend the other Research Day events. For the first time, we are offering a morning continuing education course in combination with our afternoon lectures and poster presentations. Faculty from IUSD's Center for Implant, Aesthetic and Innovative Dentistry are presenting "Integrating Contemporary Digital Technology into Your Practice" at the morning CE program.

As this year's monograph cover demonstrates the practice of dentistry as a dance of art and science. The design by Nicole Alderson, MFA, of Dental Illustrations, depicts the beauty of the shapes, forms, and colors of structures of the human body. Conversely, many works of art can be distilled to geometrical features and mathematical formulas. Dentistry is indeed a profession that combines the art of practice grounded in the science of research.

Special thanks to the Research Day Committee and the Indiana Section of the American Association of Dental Research for producing today's event. Thank you also to our event sponsors.

Very best wishes to all of the 26th Annual IUSD Research Day participants,

John N. Williams, DMD, MBA
Dean



April 09, 2018

Dear Research Day Attendees,

On behalf of the Organizing Committee and the Indiana Section of the American Association for Dental Research (IN-AADR), Uzi and I would like to welcome you to the Indiana University School of Dentistry's (IUSD) 26th Annual Research Day.

The Indiana Section is the regional link to the national (AADR) and the international (IADR) associations for dental research. The primary objective of the IN-AADR is to promote and advance basic and clinical research in all areas of the dental sciences, including the oral cavity, its adjacent structures, and the relationship to the body as a whole. Together, by utilization of this knowledge, we hope to advance science by developing new and more effective options for the prevention and treatment of oral diseases, as well as pathologies of the head and neck. Furthermore, these activities intend to foster better communication and cooperation among all – from bench researchers to clinicians – in order to share this knowledge for the benefit of all.

Within IUSD, the IN-AADR sponsors or co-sponsors invited speakers from all dental disciplines, in addition to coordinating events for the annual Research Day. By itself, Research Day offers an opportunity for investigators to present and highlight advances in basic and clinical dental research at IUSD both to their peers and in a public forum. Furthermore, Research Day offers a chance for attendees to interact with faculty and student investigators in addition to various sponsors and vendors.

This year, we are especially honored to have Prof. Raul Garcia from the School of Dental Medicine at Boston University as our keynote speaker. Prof. Garcia has made outstanding contributions to dental research in many areas including the mechanisms of periodontal disease progression, alveolar bone loss, skeletal bone mineral density, as well as tobacco control and prevention. He has also significantly advanced the areas of oral epidemiology, health services, and health policy research.

At this time, Uzi and I would like to thank all members of the Research Day Committee for their hard work and efforts toward this annual tradition at IUSD. Furthermore, we would also like to thank those who participated in the judging of research. And, last but not least, we want to thank all of you who are attending IUSD Research Day and encourage you to review the research being conducted by your fellow peers and colleagues.

Sincerely yours,

A handwritten signature in blue ink, appearing to be "FL", with a long horizontal line extending to the right.

Frank Lippert, PhD
President, Indiana Section of AADR

A handwritten signature in blue ink, appearing to be "U. Kamal", written in a cursive style.

Uzi Kamal
President, IUSD Student Research Group

Research Day Planning Committee

Frank Lippert, Chair

Nicole Alderson
Masatoshi Ando
Angela Bruzzaniti
Tien-Min Gabriel Chu
Mark Dirlam
Ygal Ehrlich
Roxana Fuentes
Dominique Galli
Richard Gregory
Yusuke Hamada
Uzi Kamal
Sue Kelly
Lisa Maxwell
Sheryl McGinnis
Keli Seering
Achint Utreja
Jeannie Vickery
Ned Warner
John Williams
Terry Wilson

Officers

Indiana Section

American Association for Dental Research

President: Frank Lippert

Vice President: Achint Utreja

Secretary/Treasurer: Councilor: Yusuke Hamada

Councilor: Anderson Hara

Chair Research Award Judging Committee: Ned Warner

Chair Staff Award Judging Committee: Angela Bruzzaniti

Officers

IUSD Student Research Group

President: Uzi Kamal

Vice President: Robert V. Holland

Secretary/Treasurer: Alexander D. Voris

Newsletter Editor: Sharon K. Iype

Faculty Adviser: Angela Bruzzaniti

Future Research Day Event: April 8, 2019 (tentative)

**Recognizing Excellence
2018 Awards**

Dental Hygiene

Elizabeth A. Hughes Dental Hygiene Case Report Award

Undergraduate Students

IN-AADR Undergraduate Student Award

Predoctoral Dental Students

AADR Student Research Day Award

American Dental Association/Dentsply International Student Clinician Award

IN-AADR-sponsored ASDA IUSD Student Research Group Award

King Saud University Travel Award for Excellence in Preventive Oral Health Care

Cyril S. Carr Research Scholarship

Graduate Dental Students

King Saud University Ph.D. Student Travel Award

Delta Dental Award for Innovation in Oral Care Research

King Saud University Travel Award for Best Clinical Case Report

Maynard K. Hine Award for Excellence in Dental Research

Staff

IN-AADR Research Staff Award

Faculty

IU School of Dentistry Alumni Association Distinguished Faculty Award for Teaching

IU School of Dentistry Alumni Association Distinguished Faculty Award for Research

King Saud University Distinguished Research Faculty Travel Award

Poster Presentations

Presenters will be at their posters to discuss their research at the following times:

3:00 pm to 3:45 pm Odd-numbered Posters and Clinical Case Reports

3:45 pm to 4:30 pm Even-numbered Posters and Clinical Case Reports

BONE BIOLOGY

P1 Osteoclast Differentiation Is Inhibited by Neurotrophin-4. E. HICKS*, S. KAWAK, J.M. HONG, A. BRUZZANITI (Indiana University School of Dentistry)

Osteoporosis, a low bone mass disease, is the result of defects in bone homeostasis which is regulated by osteoclasts, osteoblasts and matrix-embedded osteocytes. The neurotrophins (NT) have previously been shown to be important for the function and survival of neuronal cells. There are four members of the NT protein family: nerve growth factor (NGF), brain derived neurotrophic factor (BDNF), neurotrophin-3 (NT-3), and neurotrophin-4 (NT-4). Several of the NTs have been found in serum and bone marrow. We found by real-time PCR that osteoclasts, which are derived from hematopoietic stem cells in the bone marrow, express the receptors NGFR, which can bind NT-4 as well as NGF, TrkA which binds NGF, and TrkC which binds NT-3. In this study, we examined the role of NT-4 in the differentiation and activity of osteoclasts. Murine bone marrow hematopoietic cells were isolated from 6-10 week old mice and differentiated into osteoclasts with RANKL (80ng/ml) and M-CSF (20ng/ml) and in the presence of NT-4 (0, 15, 50 and 100 ng/ml). Cells were fixed after a period of 5-7 days, stained with Tartrate-Resistant Acid Phosphatase (TRAP), and counted, noting the multinucleated cells with 3 or more nuclei. In addition, the size of osteoclasts was determined microscopically. Our data demonstrated a concentration dependent decrease in the number of osteoclasts formed in the presence of NT-4. In addition, the osteoclasts formed in the presence of NT-4 (100 ng/ml) were smaller than control osteoclasts. These findings suggest NT-4 delays osteoclast differentiation, and may have an effect on the activity of osteoclasts. In conclusion, NT-4 may be important for the regulation of osteoclast activity and consequently bone mass.

P2 Kalirin and NGF Regulate Osteocyte Dendrites in a Common Pathway. L.A. DO*, T. TERHUNE, J.M. HONG, A. BRUZZANITI (Indiana University School of Dentistry)

Osteocytes are differentiated from mature osteoblasts that become trapped in the bone matrix. Osteocytes contain dendrites that communicate with neighboring osteocytes, and with osteoclasts or osteoblasts. Disruption in osteocytes number and the dendritic network has negative effects on bone homeostasis that may lead to osteoporosis and other degenerative bone diseases. Kalirin is a GDP/GTP-exchange factor that can affect cellular differentiation, proliferation and cytoskeleton remodeling. Studies from our lab have shown that Kalirin is expressed in osteoclasts, osteoblasts and osteocytes. In addition, Kalirin knockout (Kal-KO) mice have a significant reduction in bone mass. We previously found that Kalirin expression does not affect osteocyte number *in vivo*, however, Kalirin expression promote dendrite elongation of osteocytes *in vitro*. Other labs have reported that Kalirin is activated downstream of TrkA, the receptor for nerve growth factor receptor (NGF), and that NGF can promote dendrite development in neurons. We previously found that NGF promote dendrite elongation of osteocyte dendrites. For the current research, we studied whether Kalirin acts downstream of NGF to share a common pathway in regulating dendritic length. Osteocytes were isolated from Kal-KO and WT bones by collagenase digestion and cultured *in vitro*. We then treated the osteocytes with 100 ng/ml NGF for 5 days. Osteocytes were then imaged and dendrite length and number per osteocyte was calculated. Our data showed a 42.79±4.2% decrease for Kal7-KO osteocytes treated with NGF and a 39.6±4.5% decrease for Kal7-KO osteocytes without NGF treatment compared to wildtype osteocytes (p-value=0.02344, p-value=0.00618). These results indicate that Kalirin acts downstream of NFG in a common pathway to regulate dendrite length in osteocytes. In future studies, we will determine if the NFG and Kalirin is important for bone mineral density.

P3 Osteoblast-Specific Deletion of Kalirin Leads to Lower Bone Mass. D. GODFREY*, J.M. HONG, R. KOLTE, A. BRUZZANITI (Indiana University School of Dentistry)

Osteoblasts are cells on bone surfaces that are responsible for forming the fundamental structure of the bone whereas osteoclasts degrade bone. Osteocytes are derived from osteoblasts that become trapped within the bone matrix. Osteocytes communicate with osteoblasts and osteoclasts through membrane extensions known

as dendrites. Together these cells are responsible for controlling the bone mass and bone quality. Kalirin, a GDP/GTP-exchange factor, has been found to have a drastic effect on the density of bone. Previous studies showed that a global Kalirin knockout resulted in a significantly reduced bone mass phenotype. However, since Kalirin is expressed in all three bone cell types as well as other tissues, the specific role of Kalirin in osteoblasts and osteocytes is not currently known. In this study we principally examined the effects of removing Kalirin from the osteoblast lineage, which includes preosteoblasts, mature osteoblasts and osteocytes, on the bone mass phenotype of these mice. To do this, we crossed Kalirin-floxed mice with mice expressing the Cre-recombinase under control of the collagen 2.3 promoter (Col2.3) which is known to be expressed in the osteoblast lineage. The mice were selectively bred until the desired genotype was achieved and then *in vivo* bone mineral density and body composition measurements were taken with a Lunar PIXImus Densitometer. Our data shows that heterozygous Kalirin-Col2.3 knockout mice show reduced bone mass compared to control mice suggesting that Kalirin directly regulates bone mass through its actions on osteoblasts and osteocytes. Should these results continue to display significance, it is possible that further study could potentially reveal a novel target for the treatment of osteoporosis, osteoporosis, or other bone density diseases.

P4 Expression, Secretion and Function of Nerve Growth Factor in Osteoblasts and Osteocytes.

S. KIM*, S. POSRITONG, R. KOLTE, J.M. HONG, A. BRUZZANITI (Indiana University School of Dentistry)

Bone formation at dissolved bone areas, including the alveolar bone, is elicited by the recruitment of osteoblast precursors that undergo differentiation into mature osteoblasts that are capable of producing collagen. As osteoblasts mature and mineral deposition occurs, some osteoblasts become entrapped in the mineralized bone and differentiate into osteocytes. Even though several proteins involved in osteoblast function have been studied, the protein(s) that trigger osteoblast differentiation and function at damaged bone sites are still unknown. Nerve Growth Factor (NGF) is one of the neurotrophin proteins that are known to stimulate the communication, differentiation, and survival of neurons, but their role in bone cells is unclear. We examined the expression of NGF and its receptors, TrkA and NGFR/p75, in osteoblasts via real-time PCR. In addition, we cultured primary calvarial osteoblasts for 0, 4, 8, 12, 16 and 20 days in the presence of ascorbic acid and β -glycerol phosphate to induce differentiation. Cell lysates and conditioned media were analyzed by Western blotting using anti-NGF antibodies, followed by densitometry. Interestingly, NGF mRNA expression was highest in undifferentiated osteoblasts (day 0) and progressively decreased in cultures differentiated for 7, 14 and 21 days. Similarly, endogenous NGF protein levels were highest in undifferentiated osteoblasts and then declined progressively. In contrast, secreted NGF showed a biphasic response; it was high initially, decreased as the cells matured, but then increased again in late differentiation. Since late differentiation coincides with osteoblast to osteocyte differentiation, we cultured pre-osteocytic MLO-Y4 cells with recombinant NGF for 7 days and found increased mineral deposition. These findings suggest that endogenous NGF may regulate osteoblast function. In addition, secreted NGF may be involved in paracrine signaling to osteocytes to regulate cellular function, resulting in increased mineral deposition.

P5 Osteoblast Mineralization Regulated by Tyrosine Kinase Pyk2 via Estrogen Signaling.

J. WU*, P. MCINTYRE, J.M. HONG, S. POSRITONG, R. KOLTE, A. BRUZZANITI (Indiana University School of Dentistry)

Objective: Bone mass and regeneration is controlled by hormonal and cellular factors. The proline-rich tyrosine kinase 2 (Pyk2) is important for osteoblast (OB) activity, and Pyk2 knockout (Pyk2-KO) mice have high bone mass. We previously found that ovariectomized (OVX) Pyk2-KO female mice supplemented with 17β -estradiol (E2), the major female hormones controlling bone mass, have higher bone mass than wild type (WT) OVX mice. In the current study, we examined the role of Pyk2 and estrogen or raloxifene, a selective estrogen receptor modulator, on OB activity. Methods: Calvarial OBs from Pyk2-KO and WT neonatal mice were cultured for up to 28 days in osteogenic media supplemented with increasing concentrations of E2 or raloxifene. In addition, mineralization studies were performed in the presence of PHTPP, an antagonist of the estrogen receptor β (ER β). Quantitative analysis of mineral deposition was performed using an Alizarin Red-S elution assay. Results: Pyk2-KO OBs exhibited increased mineralization compared to WT OBs, which was further increased by E2 (1, 10 and 100 nM). Raloxifene (0.1 and 1.0 nM) also stimulated Pyk2-KO OB mineralization and had only a

minimal effect on WT OBs. Finally, we found that the ER β antagonist, PHTPP, decreased in a concentration-dependent manner the mineralization of Pyk2-KO OBs cultured with or without E2. Conclusions: Pyk2-KO OBs exhibit increased mineralization compared to WT OBs, which is further increased by E2 or raloxifene. Together, our studies suggest that Pyk2-deletion promotes bone formation, and increases estrogen and raloxifene stimulation of OB mineralization, by altering ER β signaling. Our studies provide insight into E2 signaling mechanisms and suggest that strategies that target Pyk2 signaling may be important for bone regeneration applications.

CARIOLOGY

P6 Fluoride's Anti-Caries Efficacy at Increasing Maturation of a Microcosm Biofilm. H. AYOUB*¹, R. GREGORY¹, E.A. MARTINEZ-MIER¹, R. LYNCH², Q. TANG³, F. LIPPERT¹ (¹Indiana University School of Dentistry, ²GlaxoSmithKline, ³Department of Biostatistics, Indiana University)

Objective: To evaluate changes in the anti-caries effect of sodium fluoride at increasing maturation stages of a microcosm biofilm grown on human versus bovine enamel. Methods: Saliva collection for this study was approved by the IRB. Three donors provided wax-stimulated saliva as the source of bacterial inoculum. A pH cycling biofilm model was modified from a published model (PMID:25576469). Three biofilm maturation stages were explored (4; 8; 12d). Microcosm biofilm was grown on 4x4mm enamel specimens (n=9 per group) for 24h using Brain Heart Infusion (BHI) media supplemented with 5g/l yeast extract, 1mM CaCl₂.2H₂O and 0.2% sucrose. The pH cycling phase consisted of daily 2x5min treatments (18.4mM sodium fluoride or deionized water), 4x15min remineralization (BHI, no sucrose, pH7.0), 3x2h demineralization (BHI, 1% sucrose, pH4.5). At the end of each maturation stage, biofilm was collected for analyses: bacterial viability (12d groups), lactic acid production (LDH), exopolysaccharide (EPS) activity. Data were analyzed using two-way ANOVA. Results: The two-way interaction for bacterial viability (12d) between substrate type and treatment type was significant (p=0.0222). For LDH and EPS analyses, the two-way interaction between the variables was significant (LDH: p<0.01, EPS: p<0.001, respectively). As the biofilm matured, both LDH and EPS activity increased significantly (p<0.05). Fluoride was able to significantly affect LDH and EPS activity at later maturation stages (8 and 12 days) [LDH: p<0.0001 (8d,human); p=0.0001 (12d,human); p=0.0016 (12d,bovine)] [EPS: p=0.0147 (8d, human); p=0.0415 (12d, human); p=0.0093 (8d,bovine); p<0.0001 (12d,bovine)]. Similarly, LDH and EPS activity were significantly different between substrate types only in later maturations (8 and 12d) [LDH: p<0.0001 (12d, treatment and control)] [EPS: p<0.0001 (8d & 12d, treatment)]. The results suggest that the anti-caries effect of fluoride increases as the biofilm matures. The substrate type influences cariogenicity of microcosm biofilm only in later maturation stages of the microcosm biofilm.

P7 Fluoride Content in Toothpastes Used by the ELEMENT Cohort. G.A. CASTIBLANCO*¹, C. BUCKLEY¹, G.J. ECKERT², F. LIPPERT¹, N. BASU³, A. BENITO⁴, A. CANTORAL⁴, A. MERCADO-GARCIA⁴, M. HERNANDEZ-AVILA⁴, H. HU⁵, B. SANCHEZ B⁶, M. TELLEZ-ROJO⁴, K. PETERSON⁶, E.A. MARTINEZ-MIER¹ (¹Indiana University School of Dentistry, ²Indiana University School of Medicine, ³McGill University, ⁴National Institute of Public Health, Mexico City, Mexico, ⁵University of Toronto, ⁶University of Michigan)

Objective: We determined total fluoride content in the five toothpaste formulations most commonly used by households with child/adolescent-mother pairs participating in cohorts of the Early Life Exposures in Mexico to Environmental Toxicants study (ELEMENT), collected as part of a cross-sectional market-basket study. Child/adolescents were 9-to-19-year-olds at the time of sample collection. We also tested toothpaste-compliance with labeled total fluoride (1450 μ g F-/g) and adherence to Mexican regulations (\leq 1500 μ g F-/g). A sample size of 34 per formulation (n=170) was determined to be sufficient to test differences (80% power, two-sided t-test, α =0.05). A study nurse visited the home of participants and transferred a sample of toothpaste from the original container to vials that were shipped to the laboratory. Toothpaste slurries (1:100 dilution) were prepared and the ADA Standard Test Method 1 (microdiffusion) and 2a (direct) were utilized for sodium monofluorophosphate- (n=1) and sodium fluoride-containing formulations (n=4), respectively. Standard curves were prepared for each test method and used for determination of the fluoride content. Fluoride was measured with an ion-selective electrode coupled to a pH/ISE meter. Descriptive statistics and two-sided t-tests (α =0.05) were performed. Results for total fluoride (μ g F-/g;mean \pm SD) in sodium fluoride formulations were: Colgate

Triple Acción (1568.4±41.5), Crest Complete (1431.1±300.4), Colgate Total 12 (1550.1±108.9), Colgate MaxFresh (1523.8±71.5), and for the sodium monofluorophosphate formulation, Colgate Máxima Protección Anticaries, (1442.3±157.6). We found that three out of four sodium fluoride formulations had fluoride levels significantly higher than stated on the label ($p < 0.05$). Furthermore, at least 59% of fluoride levels of samples from sodium fluoride and 38% of the monofluorophosphate formulations were above the allowed threshold of $\leq 1500 \mu\text{g F-g}$). Conclusion: most toothpastes used by children and families from a cohort of the ELEMENT study have total fluoride levels higher than 1450ppm. These results provide a cross-sectional estimate representative for the ELEMENT cohort.

DENTAL EDUCATION

P8 Challenges Faced by International Students in Advanced Standing Program. K. CHOPRA*, P.C. EDWARDS, M. INGLEHART (Indiana University School of Dentistry)

These programs enroll students in the second or third year of the dental school curriculum and graduates can then be licensed after graduation. There is a lack of data concerning the obstacles faced by ITD and how they deal with cultural differences, social isolation, financial issues and academic integration with traditional DDS students. The objectives are to examine these challenges relating to acceptance, opportunities, stress and discrimination from the perspective of the students enrolled in ITD Programs. Data were collected with an anonymous online survey 61 ITD students. Likert scaled and open-ended questions were used to gain a better understanding of the challenges faced by ITD students. 76 of 310 (24%) students from 9 dental schools responded to the survey. 57% reported a lack of access to academic scholarship awards, 50% financial-related stressors, 29% not being well integrated with the traditional students, 25% not having equal opportunities to participate in class governance, and 22% lack of ability to participate in national activities. Moreover, 47% experienced negative treatment due to their religious or ethnic backgrounds, 61% had concerns about the executive travel ban order, with 54% worried about its impact on the future of many Internationally Trained Dentists. The findings demonstrated that international students face quite a number of challenges during their academic years at U. S. dental schools. The most frequently named concerns were financial issues and concerns about immigration policies. Dental schools with an ITDP program should take note of these concerns and assure solid advising opportunities for their ITD students.

P9 Outcomes of a Community Engaged Oral Health Education Bilateral UK-US Exchange Program. N. LORENZANO*¹, J. BUSSARD¹, S. SCHRADER¹, J.E. KOWOLIK¹, P. WATERHOUSE², R. HOLMES² (¹Indiana University School of Dentistry, ²Newcastle University School of Dental Sciences)

The Indiana University School of Dentistry in Indianapolis, IN and The Newcastle University School of Dentistry in Newcastle, England, collaborated to create the first, bilateral civically engaged oral health education, US-UK dental student exchange program. The first two years of American dental students ($n=12$) who participated in the program were required to keep a reflective journal connecting their experiences and observations with evidenced based literature. Methods: A qualitative, thematic, social constructivist, retrospective analysis of these journals were performed by one of the participating IUSD faculty members in concert with two students who each participated as part of this program during different years. The results show that two themes and six sub-themes were found. Overall, students were greatly impacted during the exchange, by acknowledgment of their growing cultural competencies as well as overt critical recognition of their professional and personal development. Future bilateral dental student exchange programs may consider increasing the length of time abroad so as to enhance existing learning outcomes and potentially course designers may develop further curricular strategies for evaluating student outcomes. Conclusion: A bilateral student exchange which fosters interactions with local community members through civically engaged oral health care education influences a student's perspective in helping them to contextually, environmentally and socio-culturally reflect on one's own attitudes about various diverse and/or underserved people. (Supported by IUPUI Service and Learning Dissemination Grant and IUPUI Partnership Development Grant)

P10 Analyzing Student Medication Documentation in the Electronic Dental Record. W. BURCHAM*¹, E.A.S. MOSER², B.D. GITTER¹, L.M. ROMITO¹ (¹Indiana University School of Dentistry, ²Indiana University School of Medicine)

Objective: Complete, accurate documentation of patient medications facilitates appropriate treatment and helps students integrate biomedical knowledge into clinical care. This study assessed the completeness of students' medication documentation in the axiUm electronic dental record, and consistency of medications with patient-reported medical conditions. Methods: Following Indiana University IRB approval (#1503218727), 519 patient dental records were randomly chosen from axiUm query with two criteria: age > 21 years and the presence of at least one documented medication. All patient data were de-identified and records were coded. Based on medical history, each patient was assigned an American Society of Anesthesiology (ASA) Physical Status Classification System rating. Medication documentation completeness was assessed for each patient record and each medication within each record according to 6 criteria: proper medication name, class, dose/frequency, indication, potential oral effects, and correct medication spelling. Consistency was evaluated by identifying the presence / absence of a medical reason for each documented medication. Statistical analyses included frequencies and percentages, one-way ANOVA and Chi square analyses. Results: The most common correctly-recorded medication information was proper medication name (93.6%); the least common was potential oral effects (3.0%). Medication / medical condition consistency was 70.6%. Drug-level information was fully complete in 1.1% of records, and at least 70% complete in 13.7% of records. Patient-level information was fully complete in 0.2% of patients, and at least 70% complete in 6.1% of patients. Conclusion: Although consistency between patient medications and reported medical conditions was found in 7 of 10 axiUm records reviewed, students' complete documentation of each medication within patient axiUm dental records was low. This may be improved by system interventions in multiple points such as curriculum, faculty development, and clinical processes followed by reevaluation post-intervention to not only improve students' education, but their ability to treat patients more effectively.

DENTAL INFORMATICS

P11 Reliability of Patient Reported Cardiovascular Diseases Information in Electronic Dental Records. J. PATEL*^{1,3}, D. MOWERY², A. KRISHNAN¹, T.P. THYVALIKAKATH^{1,3,4} (¹Indiana University School of Dentistry, ²University of Utah, ³IUPUI School of Informatics and Computing, ⁴Center for Biomedical Informatics, Regenstrief Institute)

Cardiovascular Diseases (CVD) are the most common conditions seen in patients who are age 65 years and older. Patients with CVD need to be carefully examined as uncontrolled CVD can lead to serious consequences and adverse events. In order to know patient's accurate and up-to-date CVD status, dentists either require to rely on patients' self-reported medical histories or medical consultation. Processing medical consultations for each individual patient may not be feasible and lead to unnecessary treatment delay, pain management, time-consuming, and expensive. If patients' medical histories are accurately reported and updated then dentists can save time, cost, and manage dental diseases promptly. However, it is unclear up to what extent patients self-reported medical histories are reliable as self-reported conditions can vary based on the patient's age, gender, education level, cognitive function, income level and the presence of a number of chronic diseases. Therefore, in this study, we assessed the reliability of self-reported patients' cardiovascular diseases information stored in Electronic Dental Records (EDRs). We assessed the reliability of patients' self-reported CVD information by comparing their dental histories to their original diagnosis assigned by their medical providers in the Electronic Medical Record (EMR). To enable this comparison, we encoded patients' CVD information from the free-text data of EDRs into a structured format using advance computation methods such as Natural Language Processing (NLP). We found that our NLP approach achieved promising performance extracting patients' CVD-related information (accuracy:86% precision:98% recall:77%), we observed disagreement (Cohen's Kappa:-0.4) between self-reported EDR data physician-diagnosed EMR data. We conclude that there is an urgent need of process that can facilitate health information exchange between dental and medical providers in a timely manner from which dentists can determine patients' true CVD status in order to enable timely care.

DENTAL MATERIALS

P12 Effects of Epigallocatechin-Gallate on Dental Adhesives and MMP Inhibition. S.M. ALHIJJI*, L.J. WINDSOR (Indiana University School of Dentistry)

Matrix metalloproteinase (MMPs) have been shown to play a substantial role in dentine's collagen matrix breakdown, which may contribute to the potential failure of dental restorations over time. Therefore, modifications in the current dental adhesive system are necessary that would inhibit the uncontrolled activities of the MMPs. Epigallocatechin-3-gallate (EGCG) was selected as a MMP inhibitor to be incorporated within the dental adhesives. The main objective of this study was to investigate the potential changes in the chemical and mechanical properties of modified-dental adhesives by adding different concentrations of EGCG and evaluate their ability to inhibit MMP activity. Thus, aiming to improve the integrity and biostability of the resin-dentin interface. Disk-shaped (n=6 per group) specimens of dental adhesive (3M, Adper™ Scotchbond™ Multi-Purpose Plus) were prepared containing 0%, 0.1% (218µM), 0.05% (109µM), 0.025% (55µM), and 0.0125% (27µM) of EGCG. Gelatin zymography was used to assess each concentration of EGCG inhibitory effects on MMP-9 proteolytic activity. The degree of conversion (DC) by FTIR was utilized to relatively compare the degree of polymerization among the groups. Knoop-microhardness test was performed as an indication of any variations in the mechanical properties. Statistical analysis of variance (ANOVA) indicated no significant statistical differences among the groups in the Degree of conversion (DC) test (p=0.185), as well as Knoop-microhardness tests (p= 0.252). However, a positive correlation was found between DC and Knoop-microhardness using Pearson correlation coefficient (p<0.001). EGCG groups demonstrate a dose-dependent inhibition of MMP-9 activity by zymography. Accordingly, the incorporation of EGCG up to 218 µM did not affect the dental adhesives used in this study, and all EGCG extracts solutions showed a dose-dependent of MMP inhibition. The finding of this experimental work could potentially expand the life-span of dental restorations in the clinic by enhancing the biostability and integrity of resin-dentin interface.

P13 Effect of Chlorhexidine-Encapsulated Nanotube-Modified Adhesive on Bond Strength to Human Dentin. S. KALAGI*¹, S.A. FEITOSA¹, V. MARTINS¹, N.B. COOK¹, K. DIEFENDERFER¹, M.C. BOTTINO² (¹Indiana University School of Dentistry, ²University of Michigan School of Dentistry)

Objective: To evaluate the effect of chlorhexidine-encapsulated nanotube-modified primer and adhesive on the microtensile resin-dentin bond strength (µTBS). Materials and Methods: The primer (PR) and adhesive (ADH) from a commercially available three-step etch-and-rinse bonding system were modified by adding CHX-encapsulated nanotubes at two concentrations (CHX10% and CHX20%). The following seven groups were tested: unmodified PR+ADH (CTRL); HNT (PR+CHX-free nanotube-modified ADH); [PR-CHX10%]+ADH; [PR-CHX20%]+ADH; PR+[ADH-CHX10%]; PR+[ADH-CHX20%]; and 0.2% CHX solution. Changes in degree of conversion (%DC) and viscosity were investigated. For the µTBS, the occlusal third of human-molars were removed to expose the dentin surface. The dentin was etched with phosphoric acid, and then primer and adhesive were applied according to the aforementioned experimental groups (for the 0.2%CHX group, CHX was rubbed with a microbrush into etched dentin). The teeth were restored with resin composite. After 24h storage in distilled water, specimens were serially-sliced into sticks and then subjected to microtensile testing in a universal testing machine. Modes of failure were analyzed for all specimens. Data were analyzed using one-way ANOVA followed by Tukey's test (p<0.05). Results: When the degree of conversion of the experimental adhesives was determined with Fourier Transform Infrared Spectroscopy, there were no significant differences between any of the experimental adhesives and the control. Addition of nanotubes did not affect the viscosity of the adhesive and primer. After 24h storage, the 0.2%CHX group showed statistically higher bond strength (63.54±12.53MPa) compared to the control (47.44±13.58 MPa). The modification of the PR and ADH with CHX-encapsulated nanotubes did not affect resin-dentin immediate (24h) bond strength, as the µTBS values were similar to 0.2%CHX and control groups. Conclusion: Compared to the control (CTRL), rubbing 0.2%CHX into etched dentin increased the immediate bond strength. The long-term effect of the experimentally formulated primers and adhesives (µTBS) is currently under investigation.

P14 Fluoride Varnish Effect on Orthodontic Resin-Enamel Shear Bond Strength. B. HOAGBURG*, J.A. PLATT, S.A. FEITOSA (Indiana University School of Dentistry)

White spot lesions (WSLs) have long been a negative outcome of fixed bracket orthodontic treatment. Fluoride varnish is an effective prevention technique to reduce the incidence of WSL's. However, fluoride varnish treatment prior to resin bonding has been shown to significantly reduce resin-enamel shear bond strength (SBS). The objective of this study was to determine the effect of fluoride varnish on enamel SBS of orthodontic resin with respect to time between varnish application and resin bonding. Extracted bovine incisors were sectioned, mounted in acrylic and polished. Specimens were randomly divided into three groups (15 per group). The control group (CT) underwent a bonding protocol of prophylaxis with non-fluoridated prophylaxis paste, acid etch with 35% phosphoric acid and bonding with Reliance Light Cure Bracket Bonding Paste in a fabrication jig to create a uniform resin cylinder. The Varnish Immediate group (VI) received a thin layer of Vanish™ 5% Sodium Fluoride Varnish and 5 minutes later received the same bonding procedure as the CT group. The Varnish 1-day group (V1D) received a thin layer of Vanish™ 5% Sodium Fluoride Varnish and was stored for 24 hours, during which, the specimens were subjected to two separate 15 second tooth brushing simulations with a 1:3 aqueous slurry (Colgate Total® and deionized water) and were bonded with the same procedure as the CT group. Following bonding, specimens were stored in distilled water at 37° for 24 hours, removed and subjected to SBS testing. One-Way Analysis of Variance and Tukey tests were conducted to compare SBS among groups ($\alpha=0.05$). Preliminary results show that there was not a significant difference in SBS among the CT, VI and V1D groups. Following fluoride varnish treatment, there is not a significant decrease in SBS of orthodontic resin if enamel is bonded following manufacturer's instructions (prophylaxis, acid etch, bond).

P15 Bioactive-Resin Cement Properties and How They Interfere with Biofilm Development.

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Objective: To evaluate the mechanical and biological properties of a bioactive resin cement (ACTIVA™ bioactive- CEMENT™, Pulpdent) and how they interfere with biofilm development when compared to non-bioactive resin cements. Methods: Three resin cements were selected based on the composition and presence or absence of bioactive compounds: (AB) ACTIVA™ bioactive- CEMENT™ (Pulpdent); (EB) EMBRACE™ WetBond™ Resin Cement (Pulpdent) and (RU) Relyx Unicem 2 (3M ESPE). Film thickness, flexural strength and flexural modulus were evaluated based on ISO4049. Cytotoxicity was evaluated against fibroblast cells (NIH 3T3 cell line). Three-day-old *Streptococcus mutans* biofilm formed on resin cements specimens in the presence of 1% sucrose were examined for bacterial viability and dry-weight (n=6 discs/group). The data were statistically analyzed using One-way ANOVA followed by Tukey's test ($p<0.05$). Results: For the film thickness, all three groups passed the ISO standard. The multiple comparisons by Tukey's test revealed that bioactive resin cement (AB) presented higher film thickness than the other two resin cements (EB and RU). There was no significant difference in flexural strength among the resin cements, although the highest flexural modulus was presented by RU. The results of the cytotoxicity assay showed that, after 24 h, AC was slightly more cytotoxic than RU but not significantly different ($p>0.05$) than control. The results of colony forming units (CFU / log₁₀ CFU/ml) for *S. mutans* biofilms revealed no significant difference among the bioactive resin cement (AB) and the other two resin cements (EB and RU). Conclusion: When compared to a non-bioactive resin cement, bioactive resin cement exhibited no improvement in flexural strength, flexural modulus or inhibition of *S. mutans* biofilm formation. Like other resin cements, AB also showed no significant cytotoxic effects. Further studies are required to evaluate the role of this bioactive agent on the remineralization process.

P16 How Chlorhexidine-Encapsulated-Nanotubes Incorporated in Dentin-Adhesive Affects Biofilm Development. A.E. KARCZEWSKI*, I.E.L. VIANA, A. CARREIRO, J.A. PLATT, S. DUARTE, S.A. FEITOSA (Indiana University School of Dentistry)

Slow release of chlorhexidine (CHX) via Halloysite aluminosilicate clay nanotubes (HNT) could result in biofilm inhibition, protecting restoration marginal integrity. This study investigated the effect of chlorhexidine-encapsulated HNT-containing Scotchbond-Multipurpose dental adhesive (SBMP) on biofilm development. The groups tested were: SBMP (control), HNT (15wt.% HNT+SBMP), HNT-CHX10% and HNT-CHX20% (15wt.% CHX10%-HNT and CHX20%-HNT, respectively), CHX10% and CHX20% (15 v/v% of 10% and 20% CHX

respectively in SBMP). The HNT-free CHX groups were only analyzed with degree of conversion (DC%) and drug release tests to examine the importance of incorporating HNT. For all tests, 5x2mm discs were made. DC% was determined. To investigate CHX release, aliquots were collected after 1h, 24h and 48h, and agar diffusion sensibility test was performed. Only SBMP and HNT-containing groups were evaluated for antimicrobial efficacy. Antimicrobial sensibility was evaluated using agar diffusion against *Streptococcus mutans*. Colony-forming units (CFU) per milliliter measurement was performed against *S. mutans* biofilm. Biofilm viability was confirmed by confocal microscopy using live/dead bacterial viability kit. Data from DC%, agar diffusion, and biofilm formation were analyzed by one-way ANOVA and Tukey test ($\alpha=0.05$). For DC%, CHX-10% was the only group < 65%. For CHX release, larger zones of inhibition (ZOI) for the non-encapsulated CHX was shown in the first 24h while HNT-CHX groups presented larger ZOI later on. No ZOI was found from the control adhesive groups. HNT-CHX10% and HNT-CHX20% were similar. Considering biofilm formation, both HNT-CHX groups presented significant log₁₀ CFU/ml reduction in comparison to the control groups. Confocal microscopy confirmed reduced biofilm development and viability in HNT-CHX groups. Dentin adhesive containing nanotube-encapsulated CHX is a promising novel material for preventing cariogenic biofilm growth. Preliminary tests show non-encapsulated CHX incorporation increased curing times, in addition to altering CHX release. Integrating antimicrobial carriers can create multifunctional dental biomaterials, providing both restorative and preventative treatments.

P17 Bulk-Fill Resin Adhesive Cementation of Glass Fiber Posts. V.M. MARTINS*, L.M. ALMEIDA, C.F. SILVA, M.S. PAULA, M.S. MENEZES, C.J. SOARES, P.C.F. SANTOS-FILHO (Federal University of Uberlandia, School of Dentistry, Uberlandia, Minas Gerais, Brazil)

Objectives: This study evaluated the adhesive bond strength of glass fiber posts cemented with bulk-fill flowable resin in endodontically treated teeth. Methods: Twenty bovine incisor roots were selected and randomly divided into 2 groups (n=10). The external surfaces of the roots were coated with molding material. The canals were prepared and posts (Whitepost n° 2, FGM) cemented with resin cement (Allcem, FGM) or bulk-fill flowable resin (Opus Bulk-fill, FGM). The roots were sectioned, resulting in two 1.0-mm thick slices from the cervical, middle, and apical root regions and submitted to push-out bond strength testing (EMIC DL 2000, São José dos Pinhais, Brazil) with load cell 50N, subjected to compression loading with a constant velocity of 0.5 mm / min in the apex / crown direction. Data were analyzed using 2-way ANOVA and Tukey test ($\alpha=0.05$). Two calibrated operators determined failure mode using a stereomicroscope at 40x magnification, with a 2.5D analysis. Results: For bond strength, there was no statistically significant difference between cement and the root thirds evaluated (p=0.536). There was a statistically significant difference (p<0.001) between root regions with significantly higher values in the cervical third when compared to the others. The bulk-fill group pattern of failure was mostly mixed with resin cement partially covering the post surface in the apical third, followed of middle and cervical thirds. The most frequent failure pattern in the resin cement group was adhesive between resin cement and root dentin in the middle third, followed of apical and cervical third. Conclusion: No differences in bond strength between materials were identified. (Support Funding Agency/Grant Number: PIVICMG2017SAU039)

P18 Effects of Etching on Sub-micron Features of eMax CAD. A. WILLARD*¹, H. AL-JOHANI¹, T.M. CHU¹, R. HOWARD² (¹Indiana University School of Dentistry, ²University of Kentucky)

The aim of this study was to use atomic force microscopy (AFM), x-ray diffraction (XRD) and scanning electron microscopy (SEM) to characterize the effects of acid etching on sub-micron features of eMax CAD. Samples of eMax CAD were cut and fired in accordance to Ivoclar's recommended protocol; polished and etched with 5% hydrofluoric acid for 20, 60 and 90 seconds respectively and observed under SEM and non-contact surface profilometry (Proscan 2000). The 20s sample group was analyzed under AFM. Another batch of samples were etched with 5% hydrofluoric acid for 5, 10, 15, 30 and 60 minutes to remove the matrix phase of eMax CAD. XRD was used to identify the isolated matrix phase. The difference in etching durations had a significant effect on surface roughness values Ra on the x-axis (p=1.07e-06) but were not statistically significant on the y-axis (p=0.309). Duration of acid etch had a significant effect on surface loss (p=1.47e-04). Under AFM, there are distinctly different phases within fired eMax CAD at the sub-micron level. High-modulus grain-like structure of about 120 nm surrounded by low-modulus matrix-like phase between grains. The observation suggests segregation during the lithium disilicate nucleation process. Following the extended period of etching, XRD

results showed minimal changes in samples after extended etching. SEM results provided further visualization of the material microstructure and changes. Measurements of average surface roughness along the x-axis were found to be statistically significant, and overall trends in the data showed an increase in surface roughness as exposure time to acid etch increased. Data suggested a plateau in the material loss after 60 seconds of etching. AFM identified a high-modulus grain-like structure of about 120 nm surrounded by low-modulus matrix-like phase. Further study is needed to quantify the kinetics of etching in eMaxCAD. (Supported IUSD SRG Student Research Grant)

P19 Effects of Intracanal Medicaments on Push-Out Bond Strength. R. YAGMOOR*, J.A. PLATT, K. SPOLNIK, G.H. YASSEN (Indiana University School of Dentistry)

Objective: To evaluate the effects of typical clinical concentration as well as low-concentration Triple (TAP) and Double (DAP) antibiotic pastes on the bond strength between various root cements and radicular dentin. Materials and Methods: Intact single-rooted human teeth (n=144) were horizontally decoronated and instrumented according to standardized protocols to a final diameter of 1.5 mm. The roots were randomized into 6 experimental groups and filled with calcium hydroxide Ca(OH)₂, 1000 mg/mL TAP and DAP, or 1 mg/mL TAP and DAP for four weeks. Untreated roots served as a control. After treatment, the medicaments were removed, by irrigation with 5 mL 17% EDTA followed by 5 mL of sterile water, and each group was divided into three subgroups receiving MTA (Proroot, Dentsply), Biodentine (Septodont), or Bioceramic (Endosequence) cement for two weeks. Then two root cylinders were obtained from each root and push-out bond strength was determined. Three-way ANOVA followed by pairwise comparisons were used for statistical analyses ($\alpha=0.05$). Results: For MTA applied in the most coronal part of the roots, 1mg/mL DAP and TAP as well as Ca(OH)₂ demonstrated significantly higher bond strength compared to the clinical concentration and the untreated control group. For Biodentine applied coronally in the roots, 1mg/mL DAP resulted in significantly higher bond strength than all other experimental groups. For Bioceramic cement applied coronally in the roots, 1mg/mL DAP offered significantly higher bond strength than all treatment groups except that treated with Ca(OH)₂. Conclusion: The use of 1 mg/mL DAP resulted in significantly higher push-out bond strength compared to the clinical concentration of TAP and DAP regardless of the type of the root cement used.

ENDODONTICS

P20 Effects of Radiopaque DAP on DPSC Proliferation and Mineralization. P. MCINTYRE*¹, J. WU¹, R. KOLTE¹, G.H. YASSEN², A. BRUZZANITI¹ (¹Indiana University School of Dentistry; ²Case Western Reserve University School of Dental Medicine)

A primary goal of endodontic regeneration is to eliminate pathogenic microorganisms while allowing endogenous cells to regenerate the dentin-pulp complex. Our objective was to investigate the proliferation and differentiation of human dental pulp stem cells (DPSCs) cultured with methylcellulose (MC) containing two radiopaque agents, 30% barium sulfate (BS) or 30% zirconium oxide (ZO), in combination with metronidazole/ciprofloxacin (DAP). The experimental groups were MC+BS or MC+ZO alone or with 1-, 5- or 10-mg/mL DAP. Controls were culture media, MC only or calcium hydroxide [Ca(OH)₂]. DPSCs were cultured with the medicaments in transwell chambers and assayed for proliferation after 3 days. Additionally, DPSCs were cultured in osteogenic media and assayed for alkaline phosphatase (ALP) activity and mineralization, which are a measure of osteogenic differentiation. Experiments were replicated ≥ 3 times and data was analyzed using two-way ANOVA. The proliferation of DPSCs was similar between Ca(OH)₂, MC+BS and MC+ZO controls, as well as all groups containing 1- or 5-mg/mL DAP, whereas DPSC proliferation was inhibited with 10-mg/mL DAP. Importantly, ALP activity was significantly increased in the MC+ZO and MC+BS controls, compared to MC only. Moreover, ALP activity was higher for MC+ZO compared to Ca(OH)₂, suggesting an increase in DPSC differentiation by this radiopaque reagent. The MC+ZO+1-mg/mL DAP group also exhibited ALP activity similar to Ca(OH)₂. Lastly, we found that mineral deposition by DPSCs was similar between the MC+ZO+1-mg/mL DAP, MC+BS+1-mg/mL DAP and the Ca(OH)₂ control. These results demonstrate that BS and ZO are not cytotoxic to DPSCs and that MC+ZO+1-mg/mL DAP is comparable to Ca(OH)₂ in terms of DPSC proliferation, ALP activity and mineral deposition. Therefore, MC+ZO+1-mg/mL DAP may be a viable and superior alternative to Ca(OH)₂ for endodontic applications as it can be viewed radiographically, and promotes canal disinfection as well as the osteogenic differentiation of DPSCs to promote further root formation.

EROSIVE TOOTH WEAR

P21 The Impact of Mouthrinses on the Efficacy of Fluoride Dentifrices. E. ALBESHIR*, A.T. HARA, N.B. COOK, F. LIPPERT (Indiana University School of Dentistry)

Toothbrushing with fluoride toothpaste followed by rinsing with mouthwash is a method to maintain good oral hygiene. It is unknown to what extent these rinses can modulate the effect of fluoride in its ability to prevent erosion/abrasion. The aim of this study was to investigate and compare the impact of chlorhexidine (CHX), essential oils (EO) and cetylpyridinium chloride (CPC) mouthrinses on erosive tooth wear protection afforded by fluoride toothpastes. Methods: The following factors were considered: five rinses: CHX, EO, CPC, a fluoride rinse, and deionized water, two fluoride toothpastes: stannous fluoride (SnF₂) or sodium fluoride (NaF) and two models: (erosion/erosion+abrasion). Slabs of bovine enamel and dentin were embedded in resin blocks (n=8 per group). Specimens were subjected to a five-day cycling regimen consisting of twice-daily treatments, with or without abrasion, with fluoride toothpaste (1min) followed by mouthwash exposure (1min). Erosion (0.3% citric acid, pH2.6) was performed 5x/d. Specimens were exposed to artificial saliva during remineralization periods. Surface loss (SL) was determined using non-contact profilometry. Data were analyzed using ANOVA ($\alpha=0.05$). Results: There was no interaction among the three factors (type of toothpaste, mouthrinse and abrasion or not (dentin $p=0.0520$, enamel $p=0.4720$). There were no significant two-way interactions as SL was only affected by toothpaste and mouthwash. NaF caused less SL than SnF₂ (4.60 vs. 5.83 μm ; $p<0.0001$) in dentin, whereas the opposite was found in enamel (5.20 vs. 3.56 μm ; $p<0.0001$). Toothbrushing abrasion caused comparatively more SL in enamel (6.53 vs. 2.23 μm ; $p<0.0001$) than in dentin (6.06 vs. 4.38 μm ; $p<0.0001$). None of the tested mouthwashes affected SL. Conclusion: Commonly used mouthrinses containing antimicrobial agents or additional fluoride, do not impair the erosion/abrasion protection afforded by fluoride toothpastes. Clinical significance: The understanding of the interaction between commonly used rinses and fluoride dentifrices will help dentists provide better recommendations to patients with erosive lesions.

P22 The Impact of Age on Dental Erosion Susceptibility and Prevention. A.A. ALGARNI*^{1,2}, F. LIPPERT², P. UNGAR³, G.J. ECKERT⁴, C. GONZÁLEZ-CABEZAS⁵, J.A. PLATT², A.T. HARA² (1Taibah University College of Dentistry, 2Indiana University School of Dentistry, 3University of Arkansas, 4Indiana University School of Medicine, 5University of Michigan School of Dentistry)

This study investigated the impact of tooth age on dental erosion susceptibility and efficacy of anti-erosion preventive treatments. Extracted premolars were selected and had their age estimated (~10-100 years old) using established dental forensic methods. Enamel and root dentin slabs were obtained, embedded in acrylic blocks, flattened and polished. The specimens were assigned into one of three treatments (n=93): Sn+F (800 ppm Sn as SnCl₂; 250 ppm F as NaF, pH 4.5), NaF (250 ppm F, pH 4.5) or de-ionized water (DIW, negative control). Each was subjected to a daily cycling protocol consisting of 5-min erosive challenge (0.3% citric acid, pH 2.6) and 60-min remineralization in artificial saliva (AS), 6x/day. A 2-min treatment with the test solutions was performed at the middle of the 1st, 3rd and 6th remineralization episodes. Specimens were kept in AS overnight. Surface loss (SL) was measured after 3, 5, and 10 days, using optical profilometry. Effects of tooth age, anti-erosive treatment and time on SL were evaluated using linear mixed effects regression analysis. SL increased with age for all substrate-treatment-time combinations ($p<0.0001$). Sn+F and NaF solutions provided significantly more SL reduction compared to DIW, regardless of substrate, time, or age ($p<0.0001$), with best results shown by Sn+F solution. Tooth age did not affect the efficacy of either Sn+F or NaF solutions on enamel. For dentin, increased efficacy with age was observed at 5 (for Sn+F) and 10 days (for Sn+F and NaF solutions). In conclusion, susceptibility of enamel and root dentin to demineralization increased with tooth age. F-treatment efficacy seemed to improve with tooth age, specifically after advanced erosion simulation in dentin. Finally, Sn+F treatment effectively prevented erosion regardless of tooth age, especially for enamel substrate.

P23 Influence of Toothpaste Fluoride Concentration and Remineralization Time on Erosive Tooth Wear. S. BUEDEL*, F. LIPPERT, G.J. ECKERT, A.T. HARA (Indiana University School of Dentistry)

Remineralization of eroded dental surfaces can decrease their susceptibility to toothbrushing abrasion; however, the influence of dentifrice fluoride compound and concentration on this protection is unknown. The objective of this in-vitro study was to determine how toothbrushing of eroded teeth with fluoridated dentifrice slurries (0ppmF, 1100ppmF as NaF, 1100ppmF as SnF₂, 5000ppmF as NaF) after different remineralization times (0, 30, and 60

min) impacted the development of erosive tooth wear lesions. Enamel and root dentin specimens were prepared from bovine incisors (n=8) and submitted to a cycling protocol including erosion, remineralization at the test times, and toothbrushing, for five days. Dental surface loss (SL, in micrometers) was determined by optical profilometry. Data were analyzed using mixed-model ANOVA and Fisher's PLSD tests ($\alpha=0.05$). For dentifrice comparison, 1100ppmF as SnF₂ had significantly less SL than 0ppmF, 1100ppmF as NaF, and 5000ppmF as NaF (all $p<0.05$), regardless of substrate or remineralization time. 0ppmF neither differed from 1100ppmF as NaF ($p>0.05$) nor 5000ppmF as NaF ($p>0.05$), except when 5000ppmF as NaF was tested on enamel substrate after 60 min remineralization ($p<0.05$). 1100ppmF as NaF did not differ from 5000ppmF as NaF, except when used on dentin substrate after 30 min remin and on enamel substrate after 60 min remin ($p<0.05$). When comparing remineralization times, for 0ppmF, regardless of substrate, and for 1100ppmF as SnF₂ on enamel, no remineralization caused significantly higher SL than 30 and 60 min ($p<0.05$); with no difference between 30 and 60 min ($p>0.05$). For 1100ppmF as SnF₂ on dentin, remineralization time had no effect ($p>0.05$). For 1100 and 5000ppmF as NaF, regardless of substrate, no clear linear trend was noted. Overall, the 1100ppmF as SnF₂ dentifrice demonstrated the greatest potential for protection against toothbrushing abrasion on eroded enamel and dentin, which was enhanced by remineralization for enamel, but not dentin.

P24 Impact of Surface Micromorphology and Demineralization on Enamel Loss Measurements by Optical Coherence Tomography. M.A. ALGHILAN^{*1,2}, F. LIPPERT¹, J.A. PLATT¹, G.J. ECKERT³, C. GONZÁLEZ-CABEZAS⁴, D. FRIED⁵, A.T. HARA¹ (¹Indiana University School of Dentistry, ²King Saud bin Abdulaziz University for Health Sciences, Riyadh, Saudi Arabia, ³Indiana University School of Medicine, ⁴University of Michigan School of Dentistry, ⁵University of California)

Background and Objectives: Erosive tooth wear is a growing condition that affects the structural integrity of the tooth and should be clinically monitored ideally by non-destructive objective methods. Cross-Polarization Optical Coherence Tomography (CP-OCT) has shown to be a promising technology. This in vitro study tested whether enamel surface micromorphology and demineralization impacts enamel thickness measurements by CP-OCT. Methods: Human enamel specimens were prepared with three surface roughness levels (very rough, rough and polished; n=10). They were submitted to sequential erosive challenges, and evaluated using CP-OCT and optical profilometry (gold-standard) at baseline, and after 60, 120, 240, 360, 480, 960 and 1440 min of erosion. The effects of roughness and demineralization on enamel loss were analyzed using ANOVA ($\alpha=0.05$). Intraclass correlation coefficients (ICCs) and Bland-Altman plots were used to evaluate inter-method agreement and intra-examiner repeatability. Results: There were no significant differences in CP-OCT measurements among roughness levels ($p=0.27$) nor significant interaction between roughness and demineralization ($p=0.49$). Among demineralization periods, there was an increasing trend of surface loss as the demineralization time increased. Mean measurements at 60, 120, 240 and 480 min were significantly lower than 960 and 1440 min; and 360 and 960 min were significantly lower than 1440 min ($p<0.05$). The overall difference (mean \pm SE, in micrometers) between CP-OCT (-38.5 \pm 0.47) and profilometry (-40.3 \pm 4.1) measurements was not significant ($p=0.7261$); however, the agreement between the methods was relatively low (ICC=0.34). The measurement error by CP-OCT was ± 0.15 mm compared to profilometry. Intra-examiner repeatability was excellent (ICC=0.98). Conclusions: Enamel thickness measurements by CP-OCT were not affected by enamel roughness. The accuracy of CP-OCT compared to optical profilometry was not sufficient to monitor the magnitude of enamel loss simulated in this study.

MICROBIOLOGY / IMMUNOLOGY / ORAL BIOLOGY

P25 Effect of Quercetin on Cariogenic Traits of Oral Bacteria. R. BANSAL^{*}, R.L. GREGORY (Indiana University School of Dentistry)

Previous work demonstrated strong inhibition of *Streptococcus mutans* biofilm formation by quercetin in a dose dependent manner. Smokers have a higher number of caries-affected teeth than do nonsmokers, but the association among quercetin, nicotine, and biofilm formation by cariogenic bacteria has not been investigated in detail. Objective: The purpose of this study was to determine the effect of quercetin, a flavanoid found in many plants and foods, with varying concentrations of nicotine on biofilm formation of cariogenic bacteria. Methodology: *S. mutans* strains UA 159, UA 130, OMZ 175, 10499, and A32-2, *Lactobacillus casei*,

Actinomyces naeslundii, and *Candida albicans* were used in the present study. The minimum inhibitory concentration (MIC), minimum bactericidal concentration (MBC), minimum biofilm inhibitory concentration (MBIC), total absorbance, and biofilm formation of the strains treated with different concentrations of nicotine (0–32 mg/ml) with and without 125 µg/ml quercetin were investigated. Growth effects were measured kinetically in microtiter plates over a 24-hour period. Biofilm formation was measured using a crystal violet dye staining assay. Results: The MBIC and MBC of *S. mutans* UA 159, UA 130, OMZ 175, 10499 and rest four species with quercetin and nicotine were 16, 8, 8, 16, and 32 mg/ml and 16, 16, 16, 32 and 32mg/ml and with nicotine only respectively, and MIC was 0.25, 0.25, 0.25, 0.25 and 32mg/ml, and 16, 16, 16, 32 and 32mg/ml with and without quercetin respectively. Biofilm of *S. mutans* strains UA 159, UA 130, OMZ 175, 10499 was significantly repressed ($p < 0.05$) by 8.0–16.0 mg/ml of nicotine with quercetin and 16.0–32.0 mg/ml of nicotine alone. Conclusion: These results suggest that quercetin represses *S. mutans* biofilm formation and biofilm metabolic activity without nicotine and with low concentrations of nicotine. This may provide evidence for the efficacy of quercetin on in oral products for smokers.

P26 The Effect of Alliin on Initial Biofilm Formation of *Streptococcus mutans*. T. DIETL*, R. L. GREGORY (Indiana University School of Dentistry)

One way to prevent dental caries, the most prevalent disease in the adult population across the world, is to inhibit biofilm formation of cariogenic bacteria and thus prevent colonization in the oral cavity. Nicotine at concentrations found in smokers significantly enhances *Streptococcus mutans*, a major etiologic agent for dental caries, growth and biofilm formation. Previous research indicated that garlic extract could inhibit the biofilm formation of *S. mutans* even in the presence of nicotine. The purpose of this study was to isolate a chemical component within garlic extract responsible for these effects. Alliin is an organosulfur compound isolated from garlic, has been shown to have anti-inflammatory and anti-microbial properties and was chosen for the assay. The evaluation of alliin was done in sterile 96-well microtiter plates, adding cultures of the UA159 strain of *S. mutans* to increasing concentrations of alliin (0-2 mg/ml) diluted in Tryptic soy broth supplemented with 1% sucrose. The plates were subsequently incubated at 37°C at 5% CO₂ to imitate the environment in the oral cavity. Spectrophotometer analysis indicated significant inhibition ($p < 0.05$) of total growth and biofilm formation at a concentration of 0.25 mg/mL alliin. The results of this study suggest that alliin could be isolated as the antimicrobial component of garlic, allowing it to be used in the oral cavity to reduce the incidence of dental caries.

P27 Effect of Caffeine and Nicotine on Acrylic Denture Resin with *Candida albicans*. A. ALFAIFI*, R.L. GREGORY, J. LEVON (Indiana University School of Dentistry)

Aim: To demonstrate the impact of a wide range of caffeine concentrations on biofilm viability, metabolic activity and planktonic growth of *Candida albicans* on acrylic denture resin material in the presence or absence of 8 mg/ml nicotine. Methods: Standardized denture acrylic resin (PMMA) samples were incubated with *C. albicans* (ATCC strain 10231) and exposed to caffeine at different concentrations (0-32 mg/ml) with and without 8 mg/ml nicotine present. The first part of the study explored the combined effect of caffeine and nicotine, by measuring biofilm formation and planktonic growth of *C. albicans* (crystal violet dye staining assay for biofilm mass) and metabolic activity (XTT assay). The second part of the study, measured the viability of caffeine/nicotine-treated *C. albicans* biofilm using spiral plating on blood agar plates. Results: The addition of 8 mg/ml of nicotine (without caffeine) lead to an approximate 50% increase of planktonic growth compared to the control (without caffeine or nicotine). There was a Significant decreased effect ($p < 0.05$) on biofilm metabolic activity of *C. albicans* in the presence of 8 mg/ml of nicotine as a result of increasing caffeine concentration (16 and 32 mg/ml). However, the addition of 16 and 32 mg/ml of caffeine with 8 mg/ml of nicotine resulted in more than a 100 fold decrease in the viability of *C. albicans* biofilm without caffeine or nicotine. The results suggest that caffeine at higher concentrations (16 and 32 mg/ml) had the greatest overall inhibition on biofilm viability and metabolic activity of *C. albicans* on acrylic denture resin in the presence of 8 mg/ml of nicotine.

P28 Fibrinogen Binding of *Streptococcus mutans* Strains in Different Nicotine Concentrations.

D.E. CABRERA*, R.L. GREGORY (Indiana University School of Dentistry)

Studies have shown that there is a strong correlation between smoking and atherosclerosis. Atherosclerosis is the narrowing of the arteries due to plaque build-up on the inner arterial walls. This plaque build-up is often associated with *Streptococcus mutans*. *S. mutans* is a type of bacterium that is found in the oral cavity and responsible for tooth decay. Mucous membrane tears in the mouth allow the bacterium to enter the blood stream and adhere to the endothelial cell surface proteins that line the arterial walls. Nicotine is a major component of cigarette smoke and significantly enhances *S. mutans* growth and biofilm formation by upregulating various virulence factors including adhesins responsible for binding to different ligands. The main focus of my research was to evaluate the binding of *S. mutans* to one of the endothelial cell-produced proteins (fibrinogen) when the cells are grown in different concentrations of nicotine. Thirteen different serotype k *S. mutans* strains (kindly provided by Dr. Nakano, Japan) were grown in 0, 0.25, 0.50, 1.0, 2.0, 4.0, 8.0, 16.0, and 32.0 mg/mL of nicotine. Using an ELISA-based assay, the cells ability to bind to fibrinogen (1 ug/ml) was assessed. In addition, the biofilm growth for each *S. mutans* strain at these nicotine concentrations was measured using a crystal violet biofilm mass staining assay at an absorbance of 490 nm using a spectrophotometer and indicated that biofilm mass was increased between 4-8 mg/ml. The ELISA results demonstrated that many of the strains cultured in these same concentrations (4-8 mg/mL) of nicotine had significantly increased ($p < 0.05$) binding to fibrinogen compared to the 0 nicotine control. The fibrinogen-binding observed was correlated with the relative level of biofilm formation. This study provides additional evidence that the increased fibrinogen binding of *S. mutans* with increasing nicotine concentration may have a direct relationship with atherosclerosis.

P29 Evaluation of Antibiotics on Immature and Mature Endodontic Bacterial Biofilm. J. LEE*, R.L. GREGORY (Indiana University School of Dentistry)

The purpose of this research was to investigate the inhibitory effect of DAP against biofilm formation induced by immature and mature species collected from patients with necrotic pulps. Methods: The immature and mature species were collected using paper points from pediatric and adult patients, respectively, with root canal infections and cultured in brain heart infusion broth supplemented with yeast extract, vitamin K, and hemin (BHIY). The specific types of bacterial species that possibly make up each immature and mature species have not been fully determined, however, there was a higher number of *E. faecalis* in the immature preparation. Different concentrations of DAP (containing ciprofloxacin and metronidazole) in BHIY were added (0-20 mg/mL) with immature or mature bacteria and were cultured for 72 h in 96-well microtiter plates. Negative controls were prepared with the same bacterial species without DAP treatment. After incubation, a crystal violet assay was used to examine the amount of biofilm formation. Data were analyzed using two-way ANOVA. Results: Compared to the negative controls, DAP significantly reduced ($p < 0.05$) the biofilm formed by mature species at all tested concentrations, except at 20 mg/mL. Inhibitory effects of DAP against biofilm formation induced by immature species, however, showed no significant reduction. Conclusion: Our preliminary results provide insight into endodontic regeneration, which is associated with root canal infections that can be eliminated using different chemical treatments including DAP. Treatment with DAP attenuated the formation of biofilm induced by mature bacterial species.

P30 Effects of Various Serotypes of *Streptococcus mutans* Binding to Collagen. C. MCGREW*¹, R.L. GREGORY² (¹Indiana University School of Health and Rehabilitation Sciences, ²Indiana University School of Dentistry)

Streptococcus mutans is a cariogenic bacterial species because it can bind to connective proteins, such as collagen, in the oral cavity and produce lactic acid which demineralizes enamel surfaces. This characteristic of *S. mutans* is heightened in the presence of nicotine due to an increase in growth on the tooth surfaces and upregulation of virulence genes. Oral bacteria can enter the bloodstream many ways. Once oral bacteria, specifically those with the capability to bind to connective proteins, like *S. mutans*, enter the bloodstream, they can bind to connective proteins produced by and found on the extracellular membranes of endothelial cells in the blood vessel walls and begin to colonize. These colonies can be the initiation of arterial plaque formation, leading to hardening of the blood vessels, which can further develop into atherosclerosis. This study examined

biofilm growth of several strains with known genotypes for collagen binding in various concentrations of nicotine using crystal violet staining as well as collagen biofilm binding in various concentrations of nicotine using an ELISA method. There is a statistically significant increase ($p < 0.05$) in *S. mutans* biofilm growth and biofilm binding to collagen in the presence of nicotine concentrations in most strains from nicotine concentrations 2 to 16 mg/mL. This supports previous findings that nicotine increases *S. mutans* biofilm growth. It also supports our theory that there should be an increase in collagen binding if there is an increase in biofilm growth, and thus could lead to an increased risk of atherosclerosis in nicotine consumers.

P31 Effects of *N*-demethylation Nicotine Metabolites on *Streptococcus mutans* Biofilm Formation.

R.A. MCKINNEY*, R.L. GREGORY (Indiana University School of Dentistry)

Cigarette smoking, the most common form of tobacco product addiction, exposes the oral cavity of smokers to nicotine both during the act of smoking and for an extended period of time afterwards. Numerous studies have revealed a positive correlation between smoking and an increased incidence of dental caries. Nicotine has been consistently shown to enhance planktonic growth and biofilm formation of *Streptococcus mutans*, a facultative anaerobic oral bacteria that is the key contributor to the formation of dental caries, at physiologically relevant concentrations – between 0.25 and 4.0 mg mL⁻¹. The major metabolic pathway of nicotine, the *N*-demethylation pathway, has been studied in several different animal models and is generally well understood. In this pathway, nicotine is broken down into a series of intermediates ultimately producing norcotinine. The objective of this study was to identify how each compound actively broken down from nicotine through the *N*-demethylation pathway influenced the biofilm formation and the kinetic growth of *S. mutans*. The UA 159 strain of *S. mutans* was exposed to nicotine as well as the intermediates found within this metabolic pathway – norcotinine, nornicotine, cotinine, and trans-3'-hydroxycotinine – at physiologically relevant concentrations (0.25, 0.5, and 1.0 mg mL⁻¹). Each treatment was analyzed via crystal violet staining for biofilm formation and direct spectrophotometric kinetic growth analysis. Norcotinine, the final compound in the *N*-demethylation catabolic pathway of nicotine, significantly decreased ($p < 0.05$) the biofilm formation and kinetic growth of *S. mutans*. At a concentration of 0.5 mg mL⁻¹ norcotinine, biofilm formation was completely inhibited. (Supported by the IUSD Dental Student Research Group Fund)

P32 Effect of Titanium Dioxide-Containing Toothbrush on *Streptococcus mutans* Biofilm.

M. SANDERS*, R.L. GREGORY (Indiana University School of Dentistry)

Background: *Streptococcus mutans* is considered the etiological agent of dental caries. Soladey™ developed a toothbrush (Ion5) containing titanium dioxide which produces oxygen radicals capable of disrupting bacterial DNA synthesis and creating pores in bacterial cell walls and membranes. Objective: The objective was to examine the effectiveness of the titanium dioxide in the Ion5 toothbrush on *S. mutans* biofilm. Methods: Effects were assessed using viability and crystal violet staining. Tryptic soy broth supplemented with 1% sucrose (TSBS) was added to 6 well plates, inoculated with an overnight culture of *S. mutans* and incubated overnight to establish biofilm. The wells were rinsed and exposed to the toothbrush for 2, 4, 6 min or no treatment. The wells were rinsed, scraped to remove biofilm and plated onto blood agar plates to determine colony forming units. For crystal violet staining, TSBS inoculated with *S. mutans* in 96-well microtiter plates was incubated, planktonic bacteria were removed and the wells rinsed. Biofilm was treated with the toothbrush for 0, 30, 120, 240 and 360 sec. Biofilm was washed, stained with crystal violet, the stain extracted from the biofilm with 2-propanol and measured at 490 nm. Each experiment was replicated three times. Data was log transformed and analyzed by ANOVA to identify significant effects of exposure time. Hypothesis: It was hypothesized that titanium dioxide in the Ion5 toothbrush will reduce *S. mutans* biofilm formation. Results: Exposure time had a significant effect on bacterial counts ($p = 0.0323$). Bacterial counts after 6 min of exposure were significantly lower than both 2 min and no treatment control ($p = 0.0304$ and $p = 0.0056$, respectively). In addition, as exposure time increased the amount of remaining biofilm was statistically lower than the no treatment control. Conclusion: This study concludes that the Ion5 toothbrush kills established *S. mutans* biofilm, and is able to dislodge established *S. mutans* biofilm.

P33 Nicotine Increases *Streptococcus mutans* Biofilm and Arginine Negates the Effect.

D. WAGENKNECHT*¹, E.A.S. MOSER², R.L. GREGORY¹ (1Indiana University School of Dentistry, ²Indiana University School of Medicine)

S. mutans is detected in diseased cardiovascular tissues and strains expressing collagen binding proteins Cbm and Cnm invade endothelial cells *in vitro*. Nicotine (NIC) increases *S. mutans* biofilm and arginine (ARG) changes the biofilm by altering structure and adhesion of the extracellular polysaccharides (EPS). We investigated the effects of NIC and ARG on biofilm formation by *S. mutans* serotypes c and k strains (n=10, 10) genotyped for collagen binding protein genes (*cbm*, *cnm*). *S. mutans* grown in tryptic soy broth with 1% sucrose (TSBS) was seeded into 96-well plates with and without NIC (4 mg/ml) and/or ARG (10 mg/ml) and incubated 24 hrs at 37°C in 5% CO₂. Biofilm mass (crystal violet staining) and metabolic activity (XTT assay) were measured. *S. mutans* (Syto-9) and EPS (dextran-Alexa Fluor 568) mass were quantitated by confocal laser scanning microscopy (CLSM). Two-way ANOVA with interaction and random effect for multiple measurements was used for statistical analyses. Biofilm and metabolic data required a rank-transformation prior to analysis. NIC, ARG and NIC+ARG had significant effects on biofilm mass and metabolic activity (p<0.0001). Genotype, but not serotype, had an effect on biofilm mass (p=0.002 & 0.245, respectively). Both genotype and serotype affected metabolic activity (p<0.0001). Biofilm mass and metabolism were greatest in *cbm/cnm*⁺ strains. In CLSM NIC increased bacterial but not EPS mass (p=0.007 & 0.085). Genotype affected EPS production (p<0.0001); *cbm/cnm*⁺ strains produced more EPS with and without nicotine than *cbm*⁺/*cnm*⁺ (p=0.008) and *cbm*⁺/*cnm*⁻ strains (p<0.0001). ARG alone did not have an effect on bacteria or EPS mass, however, the effect of NIC on EPS was significantly suppressed in the presence of ARG (p=0.006). Nicotine significantly increased *S. mutans* but not EPS mass in biofilm. The *cnm*⁺ strains produced more EPS than *cnm*⁻ strains. Addition of ARG to toothpaste for smokers may negate NIC-enhanced biofilm production.

ORTHODONTICS / IMAGING / CRANIOFACIAL

P34 Orthodontic Residency Performance Predictors: Assessment of Scholastic and Demographic Parameters. A. GRILLO*, A. GHONEIMA, L. GARETTO, K.T. STEWART (Indiana University School of Dentistry)

Objective: Historically, contesting for a position in the dental specialty of Orthodontics has been very challenging. Orthodontic programs have relied upon numerous factors, including the Graduate Record Examination (GRE), as important screening tools in evaluating applicants. While, the GRE has proven to be a valuable indicator of performance in doctoral- and master's-level programs, there is a lack of information evaluating the connection between the GRE and orthodontic residency performance. The aim of this study was to evaluate the association between resident selection criteria and student performance in the orthodontic residency program. Methods: De-identified demographic and scholastic data of orthodontic graduates (n=70) from the Indiana University School of Dentistry (IUSD) was received from the IUSD Office of Graduate Education via the WebAdMIT admissions database. American Board of Orthodontics (ABO) written examination quintiles were provided by the ABO and added to the other collected information using Microsoft Excel. Data were analyzed using Spearman's correlation coefficients, one-way ANOVA, and multiple linear regression. Results: No associations were found with any component of the GRE, with the exception of the quantitative GRE section displaying weak significance with ABO Module 2 (rs= -0.31; p<0.05). Meanwhile, dental school GPA demonstrated weak correlations with all four ABO modules (rs= -0.29, -0.28, -0.30, and -0.27, respectively; p<0.05) and moderate correlation with overall orthodontic GPA (rs= 0.43; p<0.05). When assessing demographic factors, significant differences (p<0.05) were observed with the following groups demonstrating higher performance on certain modules of the ABO written examination: age (younger), race (Whites), ethnicity (Hispanics), and country of origin (US citizens). Conclusion: Findings suggest that dental school GPA, not the GRE, is a more valuable numerical indicator of residency performance. Additionally, several demographic parameters can help explain student achievement.

P35 Bolton Ratios in Hispanic Populations: A Retrospective Analysis. C. VANWANZEELE*, T.P. THYVALIKAKATH, A. GHONEIMA, K.T. STEWART (Indiana University School of Dentistry)

When considering final alignment of teeth during orthodontic treatment, there must be a harmonious relationship between the maxillary and mandibular teeth to obtain an ideal finish. Based on previous research studies, most natural teeth are well proportioned and fit together well. However, in approximately 5% of the general population, some sort of size discrepancy of the individual teeth exists.¹ The most common method for assessing an interarch tooth size discrepancy is the Bolton analysis.^{2,3} However, Bolton's original sample was small and may not apply to all populations. Currently, research is limited regarding interarch tooth size discrepancy as related to ethnicity. The objective of this retrospective study was to re-investigate the anterior and overall Bolton ratios for the Hispanic population. A group of 208 Hispanic patient records from the University of New Mexico Health Sciences Center Maxwell Museum of Anthropology Orthodontics Case File System (OCFS) was analyzed. A control group, consisting of 214 Caucasian patient records, closely matched for age and sex, was identified and used for comparison. Mesiodistal tooth width was measured on each set of records and anterior and overall Bolton ratios were calculated. One-sample t-tests were used to compare the obtained average anterior and overall ratios to Bolton's original standards. Data was analyzed using two-way ANOVA with factors for ethnicity, gender, and their interactions. No significant difference was found in the anterior ($P = 0.118$) or overall ($P = 0.216$) ratios in Hispanic subjects as compared to Bolton's standards. However, significant gender differences were found between males and females, with females having significantly smaller anterior ($P = 0.0094$) and overall ($P < 0.0001$) ratios as compared to those of males. The results suggest that Bolton's original standards apply to our Hispanic sample. However, significant gender differences suggest a need for further research to investigate this discrepancy.

P36 Low Magnitude Force for Orthodontic Expansion: Analysis of Bone Parameters.

R. ALRASHEED*, A. UTREJA (Indiana University School of Dentistry)

Transverse arch discrepancies in Orthodontics result from either a narrow maxilla or a wide mandible. Rapid maxillary expansion (RME) is routinely carried out to address this problem. However, the high magnitude of the force exerted by most maxillary expansion appliances has been shown to produce adverse effect on the teeth and the surrounding periodontium. The objective of this study was to analyze alveolar bone parameters using micro-computed tomography (CT) following maxillary expansion with a low magnitude force. Materials and Methods: Six-week-old Sprague Dawley rats ($n=20$) were divided into 2 groups: control (CTRL) and experimental (EXPT). For the duration of the experimental phase (21 days), an orthodontic wire delivering a low magnitude force was bonded using orthodontic resin to the maxillary first molars of the animals in the EXPT group. The CTRL groups did not receive this appliance. After 21 days, all the tissue samples were scanned for micro-CT analyses. The intermolar distances, midpalatal suture area and buccal alveolar bone parameters were compared between the groups using the non-parametric Mann-Whitney U test. Results: The intermolar distances measured at the root apex, midroot, cemento-enamel junction (CEJ) and occlusal surface of the maxillary first molar were significantly greater in the EXPT group compared to the CTRL group ($P < .01$). The midpalatal suture area was significantly increased in the EXPT group compared to the CTRL group at both the maxillary first and second molars ($P < .05$). Micro-CT bone parameters including percent bone volume (BV/TV), bone surface density (BS/TV), bone surface/volume (BS/BV), trabecular thickness (Tb.Th), trabecular number (Tb.N) and trabecular separation were not different among the groups. Conclusion: Maxillary expansion with a low magnitude force led to sutural expansion and orthodontic tooth movement of the maxillary molars in the buccal direction. Alveolar bone remodeling occurred in conjunction with the tooth movement.

P37 The Effect of High Bone Mass on Orthodontic Tooth Movement. R. HOLLAND*, C. BAIN, R. ALRASHEED, A. ROBLING, A. UTREJA (Indiana University School of Dentistry)

The Low-density lipoprotein receptor-related protein 5 (Lrp5) is a co-receptor of the Wnt cell signaling pathway, that is a crucial regulator of bone homeostasis. Overexpression of Lrp5 leads to a high bone mass (HBM) phenotype in mice. As alveolar bone remodeling is an integral part of the response to an orthodontic force, the **objective** of this study was to analyze the effect of Lrp5 overexpression on orthodontic tooth movement (OTM) in Lrp5-HBM mice. Materials and Methods: Two genetic variants of the Lrp5-HBM mice (A214V and G171V) and C57BL/6 wildtype mice were included in this study ($n=16$ /group). The maxillary first molar on the right

(experimental) side was tipped mesially by applying an orthodontic force with a closed-coil NiTi spring for 3 weeks. The tissues were then scanned for micro-CT analyses and processed for histology. Immunostaining for Lrp5 and sclerostin (Sost), and tartrate resistant acid phosphatase (TRAP) staining for osteoclasts were performed. The results were quantified in a predefined region of interest (ROI) in the furcation area of the maxillary first molar, and compared statistically between the groups. Results: Micro-CT analyses showed a significant decrease in the rate of orthodontic tooth movement in the Lrp5-HBM mice compared to the wildtype group ($P < .05$). Both HBM groups had higher percent bone volume (BV/TV), lower bone surface/volume ratio (BS/BV) and increased trabecular thickness (Tb.Th) compared to the WT group ($P < .05$). Histological analysis showed increased Lrp5 immunostaining in the PDL in both HBM groups compared to the WT group. Sclerostin expression was increased overall and decreased on the tension side in both HBM groups. Conclusions: Overexpression of Lrp5 decreases the rate of OTM in an animal model. Understanding the Wnt signaling pathway components involved in OTM can lead to more predictable orthodontic treatment outcomes in the future.

P38 Airway and Dentoskeletal Changes after Bone-Borne and Tooth-Borne Maxillary Expansion.

G. KAVAND*, K. KULA, K.T. STEWART, A. GHONEIMA (Indiana University School of Dentistry)

Objective: Rapid maxillary expansion (RME) is used to expand the maxilla in patients with transverse deficiency. Bone-borne RMEs were recently proposed to maximize the skeletal changes and minimize the side-effects of tooth-borne RMEs. This study compared the changes in upper airway volume after expansion with bone-borne and tooth-borne appliances. The secondary purpose was to evaluate the dentoskeletal effects of each expansion modality. Methods: This retrospective study analyzed the cone beam computed tomography images obtained before treatment and after a three-month retention period of subjects 8 to 15 years of age, who received bone-borne or tooth-borne RME. Thirty-six subjects were included in this study, 18 in the tooth-borne RME group and 18 in the bone-borne RME group. After reliability assessment, the volumetric variables and expansion measurements, obtained before and after each expansion modality, were compared using Wilcoxon signed-rank tests. Results: Intraclass correlation coefficients for inter-rater and intra-rater reliability measurements were all above 0.80. In the tooth-borne expansion group, nasal cavity and nasopharynx volume increased significantly by 12.5% and 21.8%, respectively ($p < 0.05$). In the bone-borne expansion group, nasal cavity and nasopharynx volume increased by 16.1% and 20.0%, respectively ($p < 0.05$). The volume of oropharynx and maxillary sinuses did not change significantly in either group. Dentoskeletal measurements showed that there was a significant increase in intermolar and maxillary width in both groups ($p < 0.05$); however, the buccal inclination of maxillary molars increased significantly only in the tooth-borne RME group ($p < 0.05$ for right and left first molars). Conclusion: In adolescent patients, both tooth-borne and bone borne RME resulted in an increase in the nasal cavity and nasopharynx volume, as well as expansion in maxillary dental and skeletal width. However, only tooth-borne expanders caused significant buccal tipping of maxillary molars.

PEDIATRIC DENTISTRY

P39 Utilization of Stainless Steel Crowns by Pediatric and General Dentists. A.M. CHADWICK*¹, J.F. YEPES¹, L.A. VINSON¹, J.E. JONES¹, Q. TANG¹, G.J. ECKERT², G. MAUPOME¹, T. DOWNEY³
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Dental decay affects 23% of US children aged two to five years and 56% aged six to eight years old¹. Children with extensive caries should be treated with stainless steel crowns; however, disparity exists between pediatric dentists and general dentists in restoration type provided. The purpose of this study was to evaluate the utilization of the stainless steel crown in pediatric dentists and general dentist through dental insurance claims. Data was obtained from a commercial dental insurance claims data warehouse that accrues claims from more than 50 dental insurance plans and multiple carriers in the U.S. The data encompassed all records between May 2004 and June 2016. The data included 107,487 general dentists and 5,395 pediatric dentists. The data records included 2,555,726 claims for restorations and 440,423 claims for SSCs. The data set included North Central, Northeastern, Southeastern, Southwestern, and Western practice districts. Pediatric dentists are more likely to place a stainless steel crown compared to the general dentist. The tooth most often to receive a SSC was a mandibular or maxillary primary second molar. Posterior teeth were more likely to receive a SSC

compared to anterior teeth. The majority of SSCs were placed in children aged six years old. Conclusions: Based on this national census sample of private dental insurance claims, pediatric dentists are more likely to restore carious primary dentition with stainless steel crowns compared to general dentists. Training at the dental undergraduate level should place greater emphasis on providing stainless steel crown experience.

P40 Where Do U.S. Pediatric Dentists Stand Regarding Molar Incisor Hypomineralization?

A. TAGELSIR*¹, J.A. DEAN¹, G.J. ECKERT², A.E. MARTINEZ-MIER¹ (¹Indiana University School of Dentistry, ²Indiana University School of Medicine)

Objective: With the lack of data from the US investigating dentists' level of perception regarding Molar Incisor Hypomineralization-MIH, this survey-based study targeted US pediatric dentists to determine their knowledge, perception and clinical management strategies of MIH. Methods: After obtaining AAPD and IRB authorizations, pediatric dentists identified from the AAPD's 2016-2017 membership directory in the Midwest were sent email invitations to participate in the study. The questionnaire, adopted from previous studies, had two main sections; demographics, educational and clinical practice information and MIH-focused questions including knowledge and perceptions of MIH's estimated prevalence, diagnosis, clinical challenges and restorative management options. Descriptive statistics and Chi-square tests were used for analysis. Alpha level<0.05 was considered statistically significant. Results: 251 out of 975 surveys were completed (response rate 26%). Nearly all participants were familiar with MIH. Most respondents observed MIH in either less than 10% or around 10-25% in their clinical practice (62% and 35%, respectively). The majority were aware of the lack of MIH prevalence data from the USA and believed that MIH is a significant clinical problem that demands further investigation (90% and 85%, respectively). Most respondents were very confident (65%) or confident (34%) in diagnosing teeth with MIH. The most cited clinical challenge in managing MIH teeth were "long-term success of restorations" (79%). Stainless steel crowns and composite resins were the most used dental material by respondents (32% and 29%, respectively). When analyzed individually, perceived MIH prevalence, diagnosis, clinical challenges, and restorative options differed significantly per certain demographics, educational and clinical practice characteristics (p<0.05). Conclusion: MIH is generally well acknowledged by respondents' pediatric dentists in the US Midwest region, with differences related to their perceptions of the condition's prevalence, as well as clinical and restorative management challenges.

P41 Comparison of Airway Management in Children Undergoing GA for Dental Treatment.

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Pediatric dentists often employ general anesthesia (GA) for dental rehabilitation of early childhood caries (ECC) and severe early childhood caries (S-ECC) that is delivered by a dentist anesthesiologist. Dentist anesthesiologists can manage GA in these patients using intubated or non-intubated airway management techniques. The purpose of this study is to compare the incidence of perioperative complications when intubated versus non-intubation GA is completed by dentist anesthesiologists in a pediatric dentistry setting. The Society for Ambulatory Anesthesia Clinical Outcomes Registry (SCOR) database was queried for data associated with GA cases delivered by dentist anesthesiologists from 2010-2016. Cases completed for comprehensive dental treatment in patients aged two to seven years old were identified and selected. Logistic regression compared intubated GA versus non-intubated GA for differences in the incidence of perioperative respiratory complications. Raw data collection resulted in compilation of 14,125 GA cases. After selection of pediatric cases according to inclusion criteria, the final analysis included 9,333 patients. Although incidence of respiratory complications overall was very low (0.57% of cases), the complication that occurred with the highest frequency was laryngospasm (0.32%). No significant association was found between intubated or non-intubated GA and incidence of respiratory events. When employing GA in pediatric patients for the completion of comprehensive dental treatment, there is no evidence that intubated versus non-intubated GA management is associated with an increased risk or incidence of perioperative respiratory complications.

P42 Evaluation of Restoration Longevity for Primary Maxillary Incisors. K. MCKENNA*¹, J.F. YEPES¹, J.E. JONES¹, G.J. ECKERT², Q. TANG¹, T. DOWNEY³, G. MAUPOME¹ (¹Indiana University School of Dentistry, ²Indiana University School of Medicine, ³P&R Dental Strategies)

Objective: Choosing a restoration for maxillary primary incisors (teeth nos. D, E, F, and G) that has durability and esthetics is a challenge for general and pediatric dentists. The three most common methods for restoring primary maxillary incisors are stainless steel crowns (SSCs), resin crowns (RCs), and pre-veneered stainless steel crowns (PVSSCs). The longevity of these three restorations is uncertain. In this study, the authors evaluated the longevity of the three most common treatment methods. Methods: Private dental insurance claims were obtained from a national data warehouse. The paid insurance claims (2004-2016) include the treatment provided, how many teeth were treated at an appointment, the patient age, and the type of dentist. Kaplan-Meier survival analysis and Cox Proportional Hazards analysis were used to analyze the claim information in the data set. Results: Type of treatment provided was significant in predicting the failure of the first placed restoration ($p < 0.0001$). All three restoration types have survival rates that are over 90%, but SSCs and PVSSCs had significantly better longevity than RCs, with no significant difference in longevity between SSCs and PVSSC. Conclusion: The primary maxillary incisors, teeth nos. D, E, F, and G, that were treated with SSCs and PVSSCs first, had greater longevity than teeth that were treated with RC.

P43 Blood Levels of Lead and Dental Caries. J. NORRIS*¹, J.F. YEPES¹, L.A. VINSON¹, H. HU², J. WU³, E. JANSEN³, K. PETERSON³, M.M. TELLEZ ROJO⁴, E.A. MARTINEZ-MIER¹ (¹Indiana University School of Dentistry, ²University of Toronto, ³University of Michigan, ⁴National Institute of Public Health in Mexico)

The purpose of this study was to determine if there is an association between blood lead levels and dental caries within a population of Mexican children. Data collected for the Early Life Exposures in Mexico to Environmental Toxicants (ELEMENT) cohort from a group of 490 children in Mexico City was analyzed. This data included age, sex, socioeconomic status, blood lead levels obtained every six months between the ages of 1 and 4 years, oral hygiene, diet, and dental caries evaluated clinically using the International Caries Detection and Assessment System (ICDAS) index when these same children were nine to eighteen years old. The mean age of participants when dental exams were completed was 14.3 years old. Association between number of cavities and blood lead levels were analyzed using a negative binomial regression model. The mean DMFS [decayed (D), missing (M), or filled (F) surfaces (S)] of the sample was found to be 5.5 $\mu\text{g/dL}$ with a standard deviation of 4.8. The geometric mean blood lead level was found to be 4.36 $\mu\text{g/dL}$ with a standard deviation of 2.0. No statistically significant correlation was found between dental caries in permanent teeth and blood lead levels when those teeth were developing. A statistically significant association was found between blood lead level and age, as well as blood lead level and grams of sugar consumed per day. Conclusion: This study shows a lack of correlation with exposure to lead between the ages of 1 and 4 years and dental caries in permanent dentition later in life. This information provides evidence other covariates, such as age and sugar consumption, are implemented in caries development. (Supported by RO1ES021446 (NIEHS): Prenatal and Childhood Exposure to Fluoride and Neurodevelopment)

P44 Frequency and Cost Associated with Prescribing Dental Radiographs in Children. M. WILLIAMS*¹, J.F. YEPES¹, B. SANDERS¹, G.J. ECKERT², Q. TANG¹, G. MAUPOME¹ (¹Indiana University School of Dentistry, ²Indiana University School of Medicine)

Objective: The purpose of this study was to evaluate the frequency and cost associated with routine dental radiograph prescriptions in a large sample of pediatric and general dentists in the U.S. from January 1, 2014 to December 30, 2016 and determine a cost-benefit of radiographic sequencing. Methods: Data were obtained from a commercial dental insurance claims data warehouse that accrues claims from 50+ dental insurance plans and multiple carriers in the U.S. Data extraction encompassed a 5% random sample of all records between January 1, 2014 and December 31, 2016; it included 155,542 general dentists and 5,650 pediatric dentists. Records included 4,059,952 claims for nine CDT codes of radiographs and only paid claims. We focused on the first billed and paid claim with radiographs and compared that to a Usual and Customary Rate in US Dollars per radiograph. Crucial endodontic and surgical codes were also obtained to rule out radiographic prescription due to acute treatment courses (for example, endodontic therapy or abscess drainage). Results:

Preliminary results for routine dental radiograph prescriptions showed that General Practitioners (GPs) prescribed more radiographs than Pediatric Dentists (PDs) during routine exams for ages younger than six, 10, and >12. The largest discrepancy between GPs and PDs for radiographs during routine exams was for ages younger than six, seven, eight, and 15. Conclusion: There was generally good adherence to current ADA endorsed radiographic guidelines. Prescription of radiographs by age suggests a difference between GPs and PDs.

P45 Pediatric Phantom Dosimetry Using a Hand-Held Portable Dental Radiology Device. Z. BOZIC*, J.F. YEPES, J.E. JONES, B. SANDERS, L.A. VINSON (Indiana University School of Dentistry)

Portable hand-held x-ray units are increasing in popularity as an alternative to traditional, fixed x-ray units in pediatric dental practices. The primary aim of this study was to evaluate the effective dose of exposing a pediatric phantom to bitewing and anterior occlusal radiographs using the NOMAD Portable Radiology Unit as well as to determine the amount of backscatter radiation affecting the operator. Research has shown that dosimetry, the measurement of the absorbed dose of radiation, is best studied by use of a phantom unit, which is anatomically equivalent to a human in tissue size and thickness. For this study, dosimetry was acquired using a tissue equivalent pediatric phantom simulating the anatomy of a 10-year-old child. Strategically placed dosimeters tested the absorbed dose of each image taken by the NOMAD portable radiology unit, which was maintained at a controlled angulation for the experiment in order to insure more accurate data. An additional 2 dosimeters were placed on the operator in order to record possible backscatter radiation: (1) on the operator's forehead; (2) on the operator's hand. Preliminary results indicate the NOMAD Portable Radiology Unit provides lower radiation doses to both the patient and the operator when compared to other, more traditional methods of taking radiographs.

PERIODONTICS

P46 Laser Training and Education in Postgraduate Periodontics Programs in North America.

A. RICKER*, S. PRABHU, S. BLANCHARD, D. SHIN, Y. HAMADA (Indiana University School of Dentistry)

The application of various lasers, such as Nd:YAG, Er:YAG, diode, and carbon dioxide (CO₂) lasers, to treat periodontal and peri-implant diseases is gaining momentum. While laser research is ongoing, it is important to determine if current periodontal training programs are keeping pace with these new treatment modalities and actively incorporating them into their training curriculum. The objectives of this study were to 1) determine the prevalence of laser training in North American periodontal residency programs 2) determine what types of laser devices are used 3) determine for what surgical indications each laser type is used. Following IRB approval, an anonymous electronic survey was sent to the program directors of each of the 61 periodontal residency programs. Thirty (49.1%) of the programs responded. Of these programs, 76.7% reported providing clinical training in lasers, with the diode laser being the most frequently used laser (65.2%), followed by CO₂ lasers (39.1%), Nd:YAG (26.1%), and Er:YAG lasers (26.1%). The most common procedures performed using the laser during clinical training were soft tissue surgeries, including frenectomies (91.3%) and gingivectomies (82.6%). For programs that do not currently use lasers as part of regular patient care, directors reported cost (42.9%) and lack of evidence to support its use (57.1%) as major reasons. Three out of seven programs that do not currently use lasers plan to provide laser training in the future, and 0% of programs stated that they would not provide laser training. Of the 30 programs that responded, 56.7% of program directors did not think that lasers would become the standard of periodontal/implant care within the next ten years. Laser training and education in postgraduate periodontal programs is still limited, and the majority of periodontal residents are not exposed to many types of laser devices.

P47 The Effect of Botox in Treating Gummy Smile. L. MOKEEM*, A. AL-FOUZAN, R. AL-SAQAT, A. AL-SAMARY (College of Dentistry, King Saud University, Saudi Arabia, Derma Private Clinics, Riyadh, Saudi Arabia)

Aim: To evaluate the effect of botulinum toxin A injection in improving the gummy smile level. **Materials & Methods:** This study is an experimental *in-vivo* study conducted at a dermatology clinic in Riyadh. The study includes the patients ranges from 20 to 50 year old (n=23) who received Botox injections due to excessive maxillary gingival display, excluding the patients with short clinical crowns or long maxilla, pregnant or breast feeders and patients with neuromuscular disorders. Photos were taken for the patient's smile before and after receiving the Botox injection, the photos were inserted in SketchUp program to calculate the amount of improvements in gingival display. Distance from the lower margin of upper lip to gingival margin of upper incisor was measured before and after receiving Botox. Amount of improvement was calculated, mean and standard deviation of total cases were figured out using SPSS 22.0 software system. **Results:** A total of 23 female patients received the treatment to improve their gummy smile. The level of improvement was clear 2 weeks after Botox injection. The mean percentages of improvement in the gingival display was 127% and standard deviation 78%. **Conclusion:** Botulinum toxin type A (Botox) is a temporary effective safe conservative technique according to the present sample in improving gummy smile caused by muscular hyperfunction.

P48 A New Model to Induce Chronic Experimental Periodontitis in Dogs. P. SORKHDINI*¹, N. MOSLEMI², S.H. JAMSHIDI³, A. AMIRZARGAR⁴, R. FEKRAZAD⁴ (¹Indiana University School of Dentistry, ²Tehran University of Medical Sciences School of Dentistry, ³Faculty of Veterinary Medicine, University of Tehran, ⁴AJA University of Medical Sciences)

Objective: There are several methods for induction of Chronic Experimental Periodontitis (CEP). The most common method is placing a ligature in the gingival sulcus to enhance plaque formation. However, the ligature might be missing, that results in more repeated general anesthesia needed to tie back the ligature. The objective of this study was to develop a new CEP model in dogs by using a silk ligature without the need for ligature replacement. The periodontal parameters, including Periodontal Probing Depth (PPD), Clinical Attachment Level (CAL), bleeding on probing, and redness were measured at baseline. Silk ligatures were placed around 20 teeth in each dog and fixed by using composite as an overhang material. All animals received water and a soft, plaque-promoting diet with no oral hygiene procedures. At the end of day 40, silk ligatures were removed and CEP was allowed to progress for an additional 20 days. At day 60, all dogs received scaling and root planning. No lost ligatures were observed during the 40 days that ligatures were twisted. After 60 days CEP induction period, the mean amounts of PPD and CAL increased from 1.19±0.65 mm and 0 at baseline to 2.17±0.92 and 2.23±1.36 (P<0.001), respectively. The crevicular levels of two pro-inflammatory cytokines, interleukin-1b and tumor necrosis factor- α , were evaluated at day 60. The amount of the cytokines at baseline were not measured; however, since the amount of cytokines decreased significantly after treatment, it could be concluded that this method is sensitive enough to make a significant change in cytokine profiles. This method leads to significant increases in all clinical parameters compared to the baseline. The results suggest that, twisted silk ligatures that were fixed with composite maintained their place for a long time and can be a reliable and effective model for CEP induction. (Supported by a grant from the Laser Research Center of Dentistry, Tehran University of Medical Sciences number: 88-04-97-9743)

PROSTHODONTICS

P49 Cementation, Cement Type and Vent-Holes Effect on Zirconia Copings Fit. A. MAJEED-SAIDAN*, S. HABIB, A. ALHOSSAN, M. ALQAHTANI (King Saud University, College of Dentistry)

Objective: The purpose of this in vitro study was to investigate the effect of cementation, cement type and vent holes on the marginal and internal fit of Zirconia (Zr) Copings. **Materials and Methods:** 100 extracted premolars were mounted in resin and prepared for Zr crowns. Samples were randomly divided into 5 groups (n=20): A=No Cementation; B=Cementation with Glass-Ionomer (GI); C=GI+Vent-Holes; D=Cementation with Resin Cement (RC); and E=RC+Vent-Holes. The copings were fabricated using Zirconia Blocks (ZenostarT, Wieland Dental, Germany) with a standardized cement space of 20 μ m. For Groups C & E copings were designed to have 0.5mm of Vent-Holes on the Buccal and Lingual Cusp Tips. The copings were tried on, cemented under load of 20 Newtons, thermocycled, re-embedded in resin and cross sectioned into mesial and distal halves. The copings

were examined with 3D-Digital Microscope (HIROX, KH-7700, Tokyo, Japan) at 50X to 200X magnification and the gaps or misfit were recorded at 9 predetermined points. Results: Mean gap recorded for non-cemented copings was statistically less than that for the cemented groups $P < 0.05$ except vs Group E (RC plus vent holes). No statistical difference was observed among the cemented groups $P > 0.05$. However, the fit of copings with vent holes was statistically better than without holes $P < 0.05$. Conclusions: Marginal and Internal gap values designed in the software programs differed than the actual values measured for the non-cemented copings. Cementation process also influenced the fit of the Zr copings, the resin cement being the more accurate. The addition of vent-holes on the occlusal surface improved the fit.

SALIVARY RESEARCH

P50 Salivary Cytokines and Toll-Like Receptors in Chronic Periodontitis. S. IYPE*, M. SRINIVASAN (Indiana University School of Dentistry)

Objectives: Periodontitis is a set of inflammatory diseases affecting the periodontium. Early detection of inflammation before tissue destruction in periodontitis can improve the overall well-being of an individual. Since periodontitis is a biofilm-mediated host response, soluble and epithelial cell-associated markers of microbial recognition such as toll-like receptors (TLR) and salivary cytokines are altered. The aims are: 1) to determine the salivary levels of pro-inflammatory Th1 (IL-12, INF-g), anti-inflammatory or Th2 cytokines (IL-4, IL-10) 2) to correlate the expression profiles of salivary epithelial cell-associated TLR-2 and TLR-4, receptors that recognize most periodontal pathogens in periodontitis with each of the cytokine evaluated in aim. Material methods: Saliva samples were collected from thirty systemically healthy non-smokers diagnosed with severe chronic periodontitis. Patients exhibiting Clinical attachment loss (CAL) of ≥ 4 mm in $\geq 30\%$ of sites will be included in the generalized severe periodontitis group. Thirty systemically healthy non-smokers with no known conditions will be recruited as control subjects. All samples will be centrifuged at 250g for 15 minutes at 4°C. The cellular sediment obtained will be reconstituted in isotonic saline supplemented with two drops of Zap-O-globin to lyse blood corpuscles and centrifuged at 1400 rpm for 10 min at 4°C. The epithelial cell enriched preparation will then be assessed by light microscopy for morphology, reconstituted in RPMI 1640 supplemented with 5% fetal bovine and with 5% DMSO and stored at -80°C until further analysis. Enzyme-linked immunosorbent assay (ELISA) and flow cytometry will be performed for sTLR-2, sTLR-4 and cytokines. Results, Data and Statistics: Measurable levels of IL-12 (pro-inflammatory), IL-4 (anti-inflammatory) concentrated IL-17 was present in healthy saliva. The concentration of sTLR-2 was significantly lower in the periodontitis saliva as compared with that in healthy saliva. Expression levels of TLR-2 mRNA was lower in salivary epithelial cells from periodontitis samples.

P51 Salivary Toll-Like Receptor 2 and 4 in Chronic Periodontitis. H. ALQALLAF*, S. BLANCHARD, Y. HAMADA, D. SHIN, R.L. GREGORY, M. SRINIVASAN (Indiana University School of Dentistry)

Chronic periodontitis is caused by an array of bacteria that accumulate on the teeth as plaque and lead to host-mediated destruction of the supporting apparatus of the teeth. The periodontal pathogens are recognized largely by toll-like receptors (TLRs) that bind conserved molecular patterns shared by microbial groups. Based on ligand preference, most periodontopathic bacteria are recognized by TLR-2 and TLR-4. In addition to the membrane-associated receptors, soluble forms of TLRs have been identified and are thought to regulate the host-microbe interactions and responses. Oral epithelial cells, which are abundantly found in human saliva, express most TLRs. We have previously reported elevated numbers of salivary epithelial cells (SECs) and increased expression of epithelial TLR-2 and TLR-4 in chronic periodontitis. Objective: The goal of this study is to investigate and correlate the expression profiles of soluble TLR-2 (sTLR-2) and soluble TLR-4 (sTLR-4) with paired epithelial cell-associated TLR-2 and TLR-4 in chronic periodontitis. Material and Methods: The study consisted of 20 patients with generalized chronic periodontitis (test) and 20 age-matched periodontally healthy (control) individuals. Unstimulated whole saliva (UWS) was collected. Salivary sTLR-2 and sTLR-4 in clarified saliva was measured by enzyme-linked immunosorbent assay and the TLR-2 and TLR-4 transcripts in salivary epithelial cells were assessed by quantitative real-time polymerase chain reaction. Results: The concentration of

sTLR-2 was significantly higher in the UWS of the chronic periodontitis group as compared with that of the control UWS. Although the concentration of sTLR4 was also higher in the chronic periodontitis group compared to that in periodontally healthy groups, it was not statistically significant. Furthermore, sTLR-2 directly correlated with the paired SEC-associated TLR-2 mRNA expression in subjects with chronic periodontitis. Conclusion: sTLR-2 and SEC expression of TLR2 could act as potential diagnostic/prognostic markers for chronic periodontitis.

P52 Age Related Changes in Salivary Toll-Like Receptor 2. S. STALLER*, M. SRINIVASAN, A. LINDSEY, P. THOMAS, E. RAMOS (Indiana University School of Dentistry)

The varied environment within the oral cavity support diverse microflora. The composition of oral flora changes with age correlating with microbial adhesion sites between the edentulous and the dentate status. Gram-positive bacteria are predominant in the infant mouth and the oral epithelial cells of toddlers exhibit higher microbial load. Host recognition of microorganisms is largely mediated by specialized proteins such as the toll-like receptors (TLR). In health, TLRs assist in maintaining a steady-state condition by restraining the host response to the commensal microbes. In addition to cell surface expression, soluble forms of TLRs have been recognized. Previously we reported that sTLR-2 is present in healthy saliva of children and adults. Further while sTLR-2 was elevated in children with dental caries, it was lower in periodontitis saliva. Functionally sTLR-2 is thought to quench microbes and microbial products in saliva. Since oral flora is known to change with age, we postulated that the sTLR-2 concentration in saliva is likely to be modulated by the changing composition of oral flora. By extension, assessment of sTLR-2 in saliva could suggest an indirect measure of changes in oral flora and susceptibility of oral diseases. The objective of this study is to investigate whether the sTLR-2 concentration in saliva exhibits an age associated change. Unstimulated whole saliva from different age groups was collected by the drooling method. Clarified saliva was assessed for sTLR-2 concentration by enzyme linked immunosorbent assay. Preliminary data suggest that sTLR-2 is lower in the adult samples than the pediatric samples. Soluble TLR-2 in saliva exhibits a decreasing trend with aging. Since TLR-2 has been shown to recognize the peptidoglycan of Gram+ bacteria as well as the lipopolysaccharide of Porphyromonas gingivalis, the most critical periodontal pathogen, it is tempting to speculate that the proportion of Gram positive oral bacteria decrease with aging.

TISSUE REGENERATION AND REPAIR

P53 Growth Factors from PRF in Periodontitis and Periodontally Healthy Subjects. J. CHANG*, S. BLANCHARD, R.L. GREGORY, L.J. WINDSOR, A. GOEL, Y. HAMADA (Indiana University School of Dentistry)

Objectives: Compare the amounts of growth factors from platelet-rich fibrin (PRF) between patients with periodontitis and periodontally healthy subjects, and evaluate the relationship between the amounts of growth factors from PRF and the white blood cell (WBC) and platelet counts, and pro-inflammatory cytokines (IL-1 β , IL-6, and TNF- α). Methods: Thirteen ml of venous blood was collected from patients with chronic periodontitis (test group) and periodontally healthy subjects (control group). Ten ml was used for preparation of PRF and 3 ml was used for a complete blood count (CBC). The serum from the centrifuged blood was tested for pro-inflammatory cytokines. The PRF clot was compressed into PRF membranes. The liquid exudates from compression of the PRF clot were collected. The membranes were incubated in 5 ml of saline and 1.6 ml aliquots were collected at 1, 24, and 72 hours. Platelet-derived growth factor (PDGF-BB), transforming growth factor beta 1 (TGF- β 1), vascular endothelial growth factor (VEGF), epidermal growth factor (EGF), and insulin-like growth factor (IGF) were quantified by ELISA from the collected samples. Results: Nine subjects for the test and ten subjects for the control group were included. CBC results indicated that the test group had significantly higher WBC. However, there was no significant correlation between the WBC counts and the amount of any of the tested growth factors released from PRF. The amount of growth factors released from PRF varied between individuals. The membranes had significantly more EGF, PDGF-BB, TGF- β 1, and VEGF compared to the exudates in both groups. However, no significant differences were found comparing EGF, IGF-1, PDGF-BB, TGF- β 1, and VEGF from the exudate and the membrane between test and control groups. Conclusion: PRF can be utilized as an autologous source of growth factors as it was not affected by the periodontal condition or the number of WBCs. (This study was supported by Young Researcher Grant of Osteology Foundation)

Clinical Case Report Presentations

Presenters will be at their posters to discuss their research at the following times:

3:00 pm to 3:45 pm Odd-numbered Posters and Clinical Case Reports

3:45 pm to 4:30 pm Even-numbered Posters and Clinical Case Reports

DENTAL HYGIENE

CC1 Caring For and Educating the Pregnant Patient. M. ABERNATHY*, S. TEAGARDIN, P. RETTIG (Indiana University School of Dentistry)

Objective: To educate the dental professional why treatment during the second trimester is the standard of care. Background: A 21-year-old female patient presented to the dental hygiene clinic in her 20th week of pregnancy with a chief complaint of needing a prophylaxis and a dental exam. Clinical Assessment: The patient presented with generalized plaque-induced gingivitis as evidenced by dark pink, spongy gingiva with rolled margins. Radiographs revealed generalized healthy bone levels as evidenced by 1-2mm from the CEJ to the crest of bone. She reported brushing one time per day and seldom flossing. She does not use a mouth rinse. Caries risk was high due to poor oral hygiene, high plaque score, and incipient lesions on the occlusal surfaces of teeth number 14 and 30. Dental Hygiene Care Plan: Patient received thorough oral hygiene instructions given specific to oral manifestations related to pregnancy. An oral prophylaxis, dental exam, and fluoride treatment were performed, and sealant placement was recommended on teeth number 14 and 30. Patient education consisted of explaining how important good oral hygiene is before, during and after pregnancy. Results: An appointment was made at a later date to place the sealants prior to entering the third trimester. The patient's gingival tissue and oral hygiene habits will be reassessed at this next visit. Conclusion: Dental treatment is best completed during the second trimester for the safety of both the fetus and mother and importance should be placed on specifically modifying patient education and oral hygiene habits to meet the needs of the pregnant patient.

CC2 Providing Culturally Sensitive Oral Hygiene Healthcare. A. RILEY* S. ARNOLD, A. MERRIFIELD, P. RETTIG (Indiana University School of Dentistry)

Objective: To discuss management of high caries risk patients who request no animal products to be used in their oral hygiene regimen. Assessment: A 59 year old Caucasian male presented to the dental hygiene clinic for an adult prophylaxis. Medical history was negative. The most significant dental finding was the patient's exposed root surfaces. Patient's oral hygiene status included a plaque score of 54%. Patient flosses seldom, brushes 1 time per day in the morning with sensitivity toothpaste, and reports he rarely uses a mouth rinse. Patient's dietary habits include frequent snacking on cariogenic items. Patient was classified as moderate caries risk after previous history of recent caries. Dental Hygiene Care Plan: The patient was recommended to increase brushing to two times per day, floss 3-4 times per week, and increase the use of anti-microbial mouth rinse to 4-5 times per week. The assessment indicated the recommendation of professionally applied fluoride after prophylaxis. When consent was requested to apply fluoride varnish, the patient asked if any animal products were included. Due to the patient's religious identification the products used by the patient must not contain animal ingredients and/or byproducts, but they may be tested on animals. Conclusion: Finding oral hygiene products containing no animal products is challenging not only for professional dental treatment, but also for home care use so that this can help the patient continue to improve their carries risk status.

CC3 Oral Hygiene Instructions for the Visually Impaired Patient. L. CARIE*, V. ALLEN, L. MAXWELL (Indiana University School of Dentistry)

Objective: To understand the challenges oral care providers face when providing effective oral hygiene instruction (OHI) for a patient who is visually impaired. Background: A 74-year-old African American male presents to the dental hygiene clinic for his three-month periodontal maintenance appointment. He had Nonsurgical Periodontal Therapy (NSPT) performed in 2012 and has been on maintenance since then. His chief complaint was "cleaning and exam". Medical History: His medical history was positive for hypertension, hyperlipidemia, chronic obstructive pulmonary disease (COPD), emphysema, oxygen, gastroesophageal reflux disease (GERD), anxiety, glaucoma, corneal transplants. He is considered legally blind and he takes multiple medications which cause dry mouth. Home Care: Currently, he brushes twice daily with a fluoride toothpaste, rinses with Listerine® once daily, and flosses twice a week. His plaque scores have consistently been in the 30-

36% range since 2012. These higher than recommended scores are contributing to this patient's poor gingival description. During OHI, we modified how we presented information to the patient by having the patient hold on to the toothbrush as we were brushing his teeth. This way he could feel the difference between his brushing and the corrected brushing method. All instructions given were modified to highlight his sense of touch not sight. The patient's wife was included in the hygiene instruction because she is his caregiver, with the intent that she can help him when needed. Conclusion: The patient's poor vision and xerostomia related to his medications are hindering his ability to effectively remove plaque. Modifications to oral hygiene instruction include allowing the patient to feel the brush his teeth and letting him touch the auxiliary aids. The end goal is for the patient's oral hygiene to improve if we consistently instruct more hands-on versus showing and telling.

CC4 Effects of Ill-fitting Restorations on Gingival Tissue. A. MARTINEZ*, XQ. SONG, L. MAXWELL
(Indiana University School of Dentistry)

Objective: Understand how ill-fitting restorations affect the surrounding gingival tissue. Background: A 36 year old female presented to the Dental Hygiene clinic for a cleaning. The medical history revealed no systemic diseases and she had her last dental cleaning 8 years ago in Nicaragua. All restorative treatment was done in Nicaragua. Assessment: The patient presents with generalized heavy subgingival calculus, plaque-induced gingivitis. Radiographically she has healthy bone levels and 4-6mm clinical attachment levels due to inflammation. She has several posterior restorations and has porcelain fused to metal crowns on #'s 8-10. The color of her gingiva is medium pink, with localized purple gingival margin on tooth #8. Radiographically, the crown on #8 appears to be 1mm closer to the alveolar bone than the crowns on #'s 9 and 10. Dental Hygiene Care Plan: Assessment, oral hygiene instructions, prophylaxis, and dental exam and tissue re-evaluation. A prescription for Peridex was given to the patient to address the inflammation around #8. The effectiveness of the Peridex will be evaluated at the tissue reevaluation. Discussion: Could the placement of the crown on #8 be the cause of the gingival variation seen in the assessment on tooth #8? Although there was an improvement in the patient's plaque-score at each appointment, the gingival tissue around #8, was still inflamed and purple three months after treatment began. Conclusion: Since the gingival tissue around #8 did not improve after the prophylactic cleaning, the patient was referred to Graduate Operative for further evaluation.

CC5 Long-Term Effects of Cocaine on the Oral Cavity. C. THOMAS*, R. YATES, N. STUMP (Indiana University School of Dentistry)

Introduction: There is documentation that a history of crack cocaine use negatively affects the oral tissues. When there is also a history of smoking tobacco, there are likely compounded negative effects. We will discuss the associations of cocaine usage with deterioration of oral tissues by looking beyond the effects of smoking tobacco. Case Summary: The occurrences of xerostomia, decay, erosion, and periodontitis have been found to be significantly higher among crack cocaine users. This patient displayed generalized mild alveolar bone loss, with eight teeth remaining, and a chief complaint of "I need to get my teeth cleaned." The patient's findings included generalized severe chronic periodontitis, generalized mild alveolar bone loss, and generalized mobility. The patient's medical history reveals a history of asthma, atrial fibrillation, chronic obstructive pulmonary disease, gastroesophageal reflux disease, hyperlipidemia, hypertension, osteoarthritis, and sleep apnea. The patient claims to have smoked a pack of tobacco cigarettes a day for the past 30 years. The patient reports heavy cocaine use for 15 years, but not for the last 10 years. Research has shown that when saliva and cocaine are mixed, an extremely acidic solution is created. This acidic environment can lead to the erosion of tooth enamel and the increase of decay causing bacteria. Additionally, a significant association has been found between the use of crack cocaine and reduced salivary flow, which can lead to rapid decay. Conclusion: This patient displayed generalized and severe damage to his oral health tissues. It can be concluded that it is likely that the combination of this patient's previous cocaine usage, current tobacco smoking habit, and several limiting medical health issues are contributing factors. Previous cocaine usage cannot be overlooked as a contributing factor.

CC6 Coping Strategies for Patients with White Coat Hypertension. A. YATES*, C. GONZALEZ, L. MAXWELL (Indiana University School of Dentistry)

Objective: To understand that white coat hypertension is a real disorder and to provide coping strategies for patients experiencing white coat hypertension in your dental chair. Background: A 57-year old female presents to the Dental Hygiene clinic for periodontal maintenance with a chief complaint of needing a cleaning. Her medical history was positive for hypertension, glaucoma, and sleep apnea. The patient's blood pressure reading was 180/100 mmHg RAS. After waiting five minutes, the patient's blood pressure was taken a second time and the resulting reading was similar to the first. The patient had a medical consult in her chart dated March 15, 2017 inquiring if her hypertension was under control. Her physician stated the hypertension was controlled with medication. The last blood pressure reading taken at the physician's office was 132/84 mmHg. The physician stated in the consult he believes these hypertension urgencies are caused by stressful situations such as dental appointments. Coping strategies were discussed with the patient including incorporating the tell-show-do technique, the use of stress balls, and listening to music. All of which can help manage episodic hypertension urgencies during treatment. Conclusion: With the addition of stress coping strategies, the patient's blood pressure reading taken at the end of her last appointment, decreased to 160/94, demonstrating in this instance, that the patient's stress could be managed effectively.

CC7 Dental Management of the Trigeminal Neuralgia Patient. M. CORNEWELL*, E. JOHNSON, P. RETTIG (Indiana University School of Dentistry)

Objective: To educate oral health professionals on effective management of patients with trigeminal neuralgia. Background: A 57-year-old female presented to the dental hygiene clinic for a routine bi-annual preventative appointment. The patient had been diagnosed with trigeminal neuralgia the week prior and was apprehensive about the implications of dental care. Assessment: The patient's medical history consists of degenerative disc disease in the neck, bursitis in both wrists, and gastroesophageal reflux disease. The patient has been a smoker for twenty years, smoking one pack every two days and presents clinically with heavy staining. Due to the patient's history of restorative work the caries risk status was set at moderate. Dental Hygiene Care Plan: An adult prophylaxis was performed utilizing hand instruments exclusively. The use of an ultrasonic scaler was avoided due to risk of triggering the neuralgia pain. The cleoid-discoid instrument proved effective in the removal of the accumulation of heavy stain, and the appointment concluded with a fluoride varnish treatment. The patient was given toothpaste for sensitivity and a fluoridated mouth rinse to use in her home care. Smoking cessation was also discussed due to staining, as well as the possibility of the patient's medications working more effectively. Evaluation: The patient was placed on a more frequent recall to aid in the management of deposit accumulation and staining. Conclusion: Promotion of health and prevention of disease is the primary role of the dental hygienist. Thus, being prepared with effective communication skills, knowledge about the individual patient's symptoms and triggers, and educating the patient on ways to care for their dentition at home are all crucial to comprehensive care for these patients.

CC8 Management of Tonsilloliths for the Oral Health Provider. S. COX*, C. SERMERSHEIM, L. MAXWELL (Indiana University School of Dentistry)

Objective: To aid the oral health provider in the identification and management of tonsilloliths (tonsil stones). Assessment: A 23-year-old Caucasian female presented to the Dental Hygiene Clinic for her six month dental hygiene appointment. Her chief complaint was feeling like something was caught in her throat. During the extraoral examination her tonsils felt enlarged, but not painful upon palpation. The intraoral exam revealed that her left tonsillar area appeared red and inflamed with yellow calcified deposits in the crevices of the tonsils. After further questioning of the patient, it was determined that the tonsil stones occur sporadically and without apparent cause, i.e. illness. Implementation: The oral hygiene instructions given to this patient stressed maintaining a meticulous oral hygiene regime that incorporated the daily use of an antimicrobial mouth rinse. The patient was instructed that if she felt comfortable doing so, she could carefully remove tonsil stones with a

cotton tipped applicator. Results: The patient returned six months later and reported less frequent occurrences of tonsil stones. The patient seems to believe that the hygiene plan had helped slow the developmental process of the tonsil stones and will continue to follow the recommendations we discussed. Conclusion: While the tonsil stones appeared less frequently, they are still an ongoing chronic issue and annoyance for the patient. For the oral care provider, understanding management strategies can potentially help patients decrease the occurrence of tonsilloliths.

CC9 Dental Hygiene Care for a Nothing by Mouth (NPO) Patient. S. SYKES*, S. RIGGLE, T. RADER
(Indiana University School of Dentistry)

Objective: To discuss oral hygiene care of a patient with a feeding tube. Background: An 84-year-old Caucasian male presented to the Dental Hygiene clinic for a three-month periodontal maintenance (recall). The medical history revealed that he presents with high blood pressure, atrial fibrillation, Parkinson's disease, osteoarthritis and silent aspiration. The patient also presented with a permanent feeding tube, tracheal tube and tremors from Parkinson's disease. Assessment: The patient presents with generalized plaque induced, marginal and papillary gingivitis as evidenced by dark pink, rolled margins and soft blunted papilla. The patient clinically presented with generalized moderate chronic periodontitis as evidenced by 5-6 mm CAL (levels) with a plaque score of 59%. The primary contributing factor to the patients' dental conditions would be that he has Parkinson's tremors and a permanent feeding tube. With this being said, his calculus accumulation is moderate to heavy every three months because of inadequate home care and xerostomia. This patient also has different bacteria because of the feeding tube which puts him at risk for dental caries and periodontal disease. DH care Plan: Due to rapid calculus build up the recommendation would a three-month periodontal recall, sulcular brushing with a powered tooth brush, rinsing with Act for Dry Mouth, Colgate Total Care toothpaste and Reach Flosser twice daily. Conclusion: A patient with a feeding tube presents with different dental needs to be addressed such as xerostomia, significant calculus build-up, periodontal disease and risk of aspiration pneumonia which must be addressed in the dental hygiene care plan.

ENDODONTICS

CC10 Surgical Management of Apical Fenestration: A Case Report. R. BIGGERSTAFF*, K. SPOLNIK, J. BRINGAS, Y. EHRLICH (Indiana University School of Dentistry)

Introduction: Apical fenestration is described as a pathologic condition characterized by the perforation of the alveolar bone plate and the overlying mucosa by the roots of the teeth. It has been shown to occur in 9% of cases and should be considered as a predisposing factor for persistent pain following conventional root canal therapy. This anatomic issue can easily be misdiagnosed. Misdiagnosis may cause mistreatment leading to continuation of chronic pain, potentially becoming persistent or intractable. It is critical to get a detailed history of the patient's pain along with an adequate clinical and radiographic exam. With an awareness of this issue, a simple surgical procedure can lead to patient satisfaction and potentially avoid chronic pain problems. This case report presents the surgical treatment of an apical fenestration of tooth #5 that was previously root canal treated. Methods: A 57-year-old patient presents with history of persistent, palpation pain in the apical area of tooth #5 following root canal therapy of #5. Apicoectomy performed and retrofill was completed using Endosequence Root Repair Material. Results: Following the procedure, healing was successful and patient became asymptomatic. Conclusion: Adequate knowledge and awareness with the aid of CBCT can help identify these select cases preventing overtreatment and further patient discomfort.

CC11 The Use of Platelet Rich Fibrin for Surgical Management of a Periapical Lesion. K. EPKEY*, K. SPOLNIK, J. BRINGAS, Y. EHRLICH (Indiana University School of Dentistry)

Introduction: PRF has recently been advocated as a healing biomaterial that may promote gingival and osseous tissue to heal more rapidly. PRF is composed of platelets, leukocytes, cytokines, and stem cells enmeshed in a fibrin network. PRF also includes glycosaminoglycans, which attract circulating cytokines. Cytokines play an integral role in the process of healing and regenerating injured tissues. According to Singh, the typical time required for complete osseous healing after periapical surgery is approximately one year. With the use of PRF, however, healing is accelerated and complete regeneration of bone can be seen in as little as 6 months. This case report describes the use of PRF to promote accelerated healing and regeneration of bone in the periapical

region after endodontic microsurgery. Methods: A 20 year old female presented with recurrent sinus tract #9. Surgical management of lesion was determined to be optimal treatment for best prognosis of tooth. PRF was prepared according to Choukren's protocol and root end resection and filling (with endodontic root repair material) performed on tooth #9 after curettage of apical lesion. DMFDB with PRF filtrate was then placed in surgical site and covered with PRF membrane. Sutures were then placed to achieve primary closure.

Conclusion: Use of PRF as an autograft material in endodontic microsurgical procedures promotes accelerated gingival and osseous healing by the slow release of growth factors. In this case, the four month follow up radiograph reveals almost complete osseous healing of the surgical defect.

CC12 Surgical Management of an Extensive Apical Lesion Using PRP: A Case Report. C. IBRAHIM*, K. SPOLNIK, J. BRINGAS, Y. EHRLICH (Indiana University School of Dentistry)

Introduction: Platelet rich plasma (PRP) is a blood derivative composed of a high concentration of platelets and growth factors such as platelet-derived growth factor (PDGF), transforming growth factor (TGF), insulin-like growth factor (IGF), and vascular endothelial growth factor (VEGF), which can regulate wound healing. The use of PRP in modern endodontic surgery has not been studied extensively, and reports of its use in through and through lesions demonstrate that it can have a favorable effect on hard and soft tissue healing, resulting in minimized postoperative discomfort and rapid recovery following periradicular surgery. Case report: A 36 year old male with a history of a traumatic injury to the maxillary anterior teeth presented for root canal treatment of #7. Initially, teeth #7 and #8 were treated with non-surgical root canal therapy. Three months later, the patient returned for an apical surgery. Blood was collected from the patient for the preoperative PRP preparation using a Harvest Platelet Concentrate System. A full mucoperiosteal flap was raised, lesion was enucleated, and root resection and fill were performed on teeth #7 and #8. PRP was used in conjunction with guided tissue regeneration to optimize healing. The patient returned 4 days later for suture removal, and mild swelling and minimal pain was noted. At 6 weeks, the soft tissues showed adequate healing without complications. During the 7-month follow-up, evidence of increased bone density and fill was noted radiographically. Conclusion: Beneficial effects of PRP in the treatment of a large periapical lesion were evidenced by quick, early healing and minimal post-operative pain.

ORTHODONTICS / IMAGING / CRANIOFACIAL

CC13 Unilateral Implant Restoration of Maxillary Lateral Incisor: A Case Report. J. ROSE*, J. HEINZ, T. BELLICCHI, K.T. STEWART, (Indiana University School of Dentistry)

Background: Maxillary lateral agenesis occurs in 2% of the population and more frequently in females. The loss of a tooth is especially significant when it occurs in the esthetic zone. Contemporary treatment options include: canine substitution via orthodontics, resin bonded bridge, fixed or removable prosthesis, or space re-distribution for a dental implant. Replacement of the maxillary lateral incisor with an implant can be quite complicated and requires coordination between multiple dental professionals. The purpose of this case report is to demonstrate the interdisciplinary treatment of a patient needing a unilateral implant in the esthetic zone. Case Information: A 33-year-4-month old female presented to the orthodontic clinic with the chief complaint: "I want to close my spaces." She presented with generalized spacing due in part to missing dentition: upper right maxillary lateral, upper left 1st molar, and lower right lateral incisor and a high smile line. The patient's diagnosis included: skeletal class II, Angle class IV malocclusion (left) and Angle class II (right) with flared maxillary incisors and protrusive lower incisors. The patient agreed to the interdisciplinary dental treatment that included comprehensive fixed orthodontics (.022 Opal brand brackets with MBT prescription) for space closure, while maintaining space for an upper right maxillary lateral incisor implant. Throughout orthodontic treatment there was periodic visits to the prosthodontist for evaluation of the implant site. Immediately following orthodontic treatment, an oral surgeon placed a 2.9mm Roxolid Straumann implant at the upper right maxillary lateral incisor position. To maintain the space and confirm the shape and color of the tooth, the prosthodontist fabricated a provisional abutment. Results: Orthodontic treatment time was 19 months and a satisfactory treatment outcome was achieved through an interdisciplinary treatment approach. Conclusion: A unilateral maxillary lateral implant can be a viable treatment option, with careful planning and cooperation between specialists.

CC14 Orthodontic and Surgical Management of a Class III Malocclusion. K. BOTSFORD*, J. LAMBERT.
C. KLENE, M. FRAZIER (Indiana University School of Dentistry)

Background: Orthodontics manages the relationship of not only the teeth but also the jaws. Sometimes, orthognathic surgery in conjunction with orthodontics is recommended to achieve an ideal facial and occlusal result. Maxillary LeFort I osteotomies are a type of surgery commonly performed to correct midface deficiencies and provide reliable long-term results. Patient Background: In this case report, a twenty-four year old, Caucasian male presented with a retrusive maxilla and class III dental malocclusion. Initial facial presentation included malar deficiency, retruded upper lip and deficient incisal display. Intraorally, there was a reverse overjet of -2mm, overbite of 0mm, and mild crowding in both arches. Maxillary and mandibular incisors were proclined and a bilateral posterior openbite of 1mm was present. Cephalometric analysis revealed maxillary skeletal retrusion and mandibular skeletal prognathism. Fixed orthodontic appliances were used to align and level the dental arches. Following nine months of pre-surgical orthodontic treatment, a LeFort I maxillary advancement and downgraft osteotomy surgery was performed. An additional sixteen months of post-surgical orthodontics was rendered for space closure and case detailing. The total treatment time was twenty six months. Results: Following comprehensive orthodontics with surgical maxillary LeFort I advancement and downgraft osteotomy, the patient achieved an Angle's class I occlusion. Ideal overbite and overjet was established. Incisal display was increased and a consonant smile arc was obtained. A significant facial improvement occurred with a more esthetic profile and lip support following treatment. Skeletal harmony between the maxillary and mandibular arches in relationship to each other and the cranial base were achieved. Conclusion: Comprehensive orthodontic treatment in conjunction with orthognathic surgery significantly improved the dental malocclusion, skeletal relationship and the facial esthetics of the patient.

CC15 Interdisciplinary Management of a Severe Class III Skeletal Malocclusion. C. BENINATI*, M. FRAZIER, C. KLENE (Indiana University School of Dentistry)

Literature Background: Orthognathic surgeries are typically used in conjunction with orthodontic treatment to improve patient's facial profile and eliminate skeletal discrepancy between the maxilla and mandible. Although a major motivating factor for orthognathic surgery may be esthetic facial changes, some authors note that, more often than not a patient who undergoes orthognathic surgery has a post-operative period of time where they have difficulty adjusting to their new appearance. Patient Background: A 23 year old female presented with a large class III skeletal malocclusion. Her chief complaint was "My jaw is sticking out too far forward and I can't speak". Her medical history included myopathy and severe difficulty speaking. Dental analysis showed an initial overjet of negative 7mm, an anterior open bite of 7mm, bilateral maxillary transverse deficiency with dental protrusion, mandibular dental retrusion, and missing teeth #14 and #19. Skeletal analysis revealed maxillary skeletal retrusion, increased mandibular length, and a hyperdivergent mandible. Treatment Results: Bilateral sagittal split osteotomy, maxillary advancement, and posterior impaction combined with orthodontic treatment yielded an adequate esthetic result. The patient's facial profile, occlusion, overjet, and overbite were improved significantly and an Angle's class I canine was achieved. Despite the improvement in facial balance, for the first 4 months post-surgery the patient repeatedly stated she felt "like a chipmunk". We observed a temporary decline in the satisfaction of her appearance post-surgically. Conclusion: This combined orthodontic and orthognathic surgical approach to treatment showed significant improvement of both the originally presented dental and skeletal discrepancies of the maxilla and mandible. The patient's speech and occlusal function were dramatically improved. There is a psychosocial aspect to the acceptance of orthognathic surgery that clinicians should remain aware of.

CC16 Treatment of Mandibular Retrognathia with Combined Orthodontic and Orthognathic Surgical Approach: A Case Report. S. RYAN*, C. KLENE, M. FRAZIER (Indiana University School of Dentistry)

Literature Background: Bilateral sagittal split osteotomy (BSSO) is a reliable procedure for advancement of the mandible to improve facial esthetics and class II malocclusion. Additional reported benefits of BSSO in the literature include airway dimensional increases and improvements in masticatory function. The procedure requires substantial planning and interdisciplinary collaboration between the orthodontist and oral surgeon. Patient Background: A healthy 14-year, 5-month-old Caucasian female presented with a severe class II division 1 malocclusion with painful deep bite with palatal impingement. Initial overjet was measured at 11mm. Initial

facial presentation included decreased lower facial height and marked mandibular retrognathia. Initial intraoral presentation included generalized 7mm maxillary spacing with a 2mm midline diastema. Cephalometric analysis revealed severely hypodivergent mandible, protruded and proclined maxillary incisors, and mandibular skeletal retrusion. Fixed appliances and a nonextraction approach were utilized with en-masse maxillary space closure and sliding mechanics. Leveling of the curve of Spee was achieved with a step in the occlusion pre-surgically. Following 2.75 years of pre-surgical orthodontics, a BSSO mandibular advancement procedure was performed. Post-surgical orthodontics required 1.25 years for completion. Treatment Results: BSSO surgery in conjunction with orthodontic treatment yielded 5mm of A-P mandibular advancement. An Angle's class I occlusion was achieved. Overjet and overbite were reduced to ideal values. An increase in chin projection and facial balance was noted. Conclusion: A combined orthodontic/orthognathic surgical approach to treatment of this patient yielded a dramatic improvement in dental, skeletal, and facial dimensions as well as quality of life.

PERIODONTICS

CC17 Amnion-Chorion Allograft Membrane: Predictable Material for Periodontal Regeneration: A Case Report. A. GOEL*, Y. HAMADA. (Indiana University School of Dentistry)

Periodontal defects are associated with deep probing depths providing an access to plaque and other bacterial products. Regenerative surgical procedures with bone graft substitutes, barrier membrane and biologic modifiers are well established. Many studies have shown that Amnion-Chorion membrane (ACM) consists of multiple growth factors such as PDGF-AA, BB, FGF, EGF, TGF- β to enhance the healing process. The aim of this case report is to present a novel approach to utilize the ACM over the root surface. A 31-year-old African American female presented to the Graduate Periodontics Clinics at IUSD for treatment of periodontal diseases on #5, 27 and 30. Radiographic finding at these sites showed intrabony defects ranging between 5-7mm. Guided tissue regeneration (GTR) with combination of ACM and Freeze-Dried Bone Allograft (FDBA) was recommended. After informed consent was obtained, in 2 subsequent appointments, #5,27 and 30 were treated in the similar manner. A full thickness muco-periosteal flap was raised both on buccal and lingual sides. Thorough debridement of the defects and root planing were done. Intrabony defect appeared to be 2 wall defects with 5-6mm vertical component on all sites. After completion of debridement, 17% EDTA was used to condition the root surface and remove the smear layer. Following irrigation with saline, ACMs were cut in half and one half was placed over the root surface. Hydrated FDBA particulate was used to fill the defect and was covered with another half of the ACM. The flaps were repositioned with 5-0 dPTFE sutures to obtain primary closure. Healing was uneventful via primary intention. Six-months after those procedures, radiographically and clinically optimum results were achieved. This report suggests that Amnion-Chorion membrane growth factors might improve the periodontal regenerative surgeries by providing a mechanical barrier effect and multiple growth factors to enhance the healing process for GTR procedures.

CC18 Management of a Recurrent Gummy Smile Case after Seven Years. Y. YEH*, S. BLANCHARD (Indiana University School of Dentistry)

Introduction: This case report describes the management of a patient with esthetic concerns of gummy smile. The patient stated that the area had been previously treated with periodontal surgery seven years earlier but has slowly recurred over time. Case Presentation and Results: A 32 year old African-American female first presented to IUSD Graduate Periodontal Clinic in 2010 with a chief complaint that she showed too much gingiva when she smiled. After evaluation, patient was diagnosed with excessive gingival display from teeth #3-14, and esthetic crown lengthening was performed. She was satisfied with the clinical outcome one week post surgically with the new gingival margin position at the level of the CEJs, but she did not return for further follow-up visits. Seven years after surgery, the patient came back for evaluation of her gummy smile again with the gingival margin approximately 3 mm coronal to the CEJs of #6-11. A second esthetic crown lengthening surgery was performed again with additional bone reduction on the facial aspect to a level 3 mm apical to the CEJs of #6-11. The additional 1 mm of bone reduction and additional osteoplasty to reduce the bone thickness was performed. Three months after the second surgical procedure, the patient was satisfied with her smiling with minimal

gingival display. The gingiva appeared healthy with full exposure of the anatomic crowns. Possible reasons for recurrence of the excessive gingival display are most likely related to: inadequate bone removal at time of the first surgery that was compounded by a thick gingival biotype. Conclusions: The amount of bone removal will be a critical point to prevent the future relapse of gummy smile and the amount of bone reduction (ostectomy) should equal the amount of additional tooth exposure that is desired to accommodate dimensions for the biologic width to minimize relapse.

PROSTHODONTICS

CC19 Utilizing Digital Option for Oral Rehabilitation of Edentulous Patient. D. DIAZ RUBAYO*, J. LEVON (Indiana University School of Dentistry)

Patient was referred from IUSD for treatment. Patient was diagnosed with Ectodermal Dysplasia, cleft lip and palate, fully edentulous, skeletal relationship class III. Goals of treatment: Correct profile, skeletal relationship with LeFort I surgery; Full arch prosthesis rehabilitation enhanced by implants for fixed or removable prosthesis. Materials: Treatment was digitally planned with triple scan technique using CBCT, 7series scanner and Dolphin for facial profile simulation, CoDiagnostiX for implant position planning, 3D printed guides with Forms2 printer. Methods: After carefully planning restoration of the patient in a wax try in with teeth set in a Class III dental relationship a dual scan CBCT was done. Dolphin was used to simulate the LeFort surgery for splint design. The three parts: upper, lower “dentures” and splint were 3D printed using the Forms2 printer and used in the LeFort surgery with Iliac crest graft. Four months after surgery Dual Scan was repeated to assess graft and plan for implant position based on the Prosthetic rehabilitation. Using CoDiagnostiX, mandibular implants were designed for a full arch fixed prosthesis while maxillary implants will be postponed due healing time of graft. Bone reduction guides and implant guide were used in surgery. Results: The interdisciplinary approach without a doubt is the best way to treat a patient that requires complex treatments like this one. Communication between departments might be challenging but with the use of the available technology, video conference and digital planning, the Prosthetic and Surgical team were able to integrate their knowledge for a better predictable outcome. Digital planning software provides a good tool for patient education and interdisciplinary communication.

CC20 Managing Esthetic Complications with Digitally Fabricated Complete Dentures. S.E. AINOOSAH*, H.D. ALSAYED, J. LEVON, D. MORTON (Indiana University School of Dentistry)

With the emergence of Computer Aided Design and Computer Aided Manufacturing technology, digital designing and milling of the removable complete denture prosthesis is highly beneficial. However, esthetic complications may occur involving the shape and/or shade of the teeth and with the amount of gingival display. A 66-year-old female patient presented with a digitally fabricated maxillary complete denture that was made 6 months ago. Her chief complaint was, “I don’t like how my upper teeth look like, my teeth are very long and I don’t show much of the gingival color.” Clinical assessments of the occlusal vertical dimension, centric relation, and phonetics revealed sufficiently fabricated dentures. However, the maxillary teeth were very long and the patient didn’t show enough of the pink acrylic resin. Since the location of the incisors edges were accurate, gingival wax up was made on the existing denture to mask the cervical areas of all the maxillary teeth and it was given to the patient overnight on a “trial” basis. After 24 hours, the patient came back happy and satisfied. The corrected maxillary complete denture was scanned and a new maxillary complete denture was milled. Finally, the denture was minimally adjusted and delivered to the patient. Patient was seen for the follow up at which time she expressed her satisfaction with the overall results. Although the process of fabricating digitally designed removable complete denture is clinically predictable, there are drawbacks that require an alternative approach to excellently satisfy patients’ treatment and desire.

CC21 Prosthodontic Management of Unfavorably Inclined Implants: A Clinical Report. A.A. AL-SHAHRANI*, H.D. ALSAYED, J. LEVON, D. MORTON (Indiana University School of Dentistry)

Implants have been used to support dental prostheses for many decades, but the use of implants in dentistry has dramatically increased in recent years. Nowadays, dental implants are becoming the norm for the replacement of missing teeth. This clinical report will describe the prosthetic management of unfavorably inclined implants. A 69-year-old female presented to the Graduate Prosthodontic Clinic at Indiana University

School of Dentistry with a chief complaint of “I want to restore my implants”. Her dental implants were placed two years ago in area # 12-13. They had osseointegrated well within the surrounding bone. However, they presented with a severe buccal inclination. In addition, the two implants were placed very deeply within the bone. Provisional crowns were fabricated with the screw access openings coming out from the buccal aspect of the provisional crowns. The prosthodontic management of these dental implants with unfavorable inclination was via the use of custom cast UCLA abutments that screwed in from the occlusal. After the fabrication of the fixed dental prosthesis, it was screwed into position and the access holes were covered with composite resin. This treatment strategy resulted in both functional and esthetic restorations which eliminated the use of cement to retain them. Patient was seen for follow up and no complications were found. Conclusion: This poster presentation demonstrates the use of custom cast UCLA abutments for the prosthodontic management of unfavorable inclined dental implants.

CC22 Rehabilitation of a Completely Edentulous Patient with Oroantral Communication: A Clinical Report. A.K. THORN*, J. LEVON, D. MORTON (Indiana University School of Dentistry)

The present clinical report describes the prosthetic rehabilitation of a patient with a maxillofacial defect of unknown diagnosis, specifically, an oroantral communication which is defined as a pathological opening between the oral and nasal cavities. One of the rehabilitation modalities for this type of defect is the non-surgical approach with a palatal bulb. In this case, such a type of obturator was used as a component of the dental prosthesis in order to fit and close the palatal defect. Case report: 58-year-old, caucasian female, who's chief complaint was based on the incapacity of talking and eating due to a complete lack of soft palate. Medical history: COPD, arthritis, inherited musculoskeletal degenerative disease, nodule at the thyroid and anxiety. Extraoral examination: bilateral angular cheilitis, nose deformity and Angle's Profile: Class I. Intraoral examination: maxillary and mandibular edentulous, House's Arch Form: Class 3, interarch space: Class I, ridge parallelism: Class I, Angle's Ridge Relationship: Class I, PDI: Class IV, bone quantity: Class A, mandibular ridge form: Class 3, Neil's Lateral Throat Form: Class 2. Treatment plan consisted of the fabrication of a maxillary removable complete dental prosthesis/palatal bulb and a mandibular removable complete dental prosthesis, which teeth were arranged in a bilateral balanced occlusion. Conclusion: Patient was extremely receptive and pleased with the esthetic and functional outcomes. More importantly, her self-esteem was improved tremendously.

TISSUE REGENERATION AND REPAIR

CC23 The Use of a CO2 Laser in Dental Implant Site Development. J. VILLANUEVA*, S. BLANCHARD, Y. HAMADA (Indiana University School of Dentistry)

The use of lasers is an increasingly attractive treatment modality in clinical dentistry. There are a number of different types of lasers used in dentistry which can be used on soft tissue, hard tissue or both. Among the commercially available lasers, the carbon dioxide (CO2) laser has been used in dentistry since the 1970's. The purpose of this case report is to demonstrate a novel approach for accelerating soft and hard tissue healing using a CO2 laser for alveolar ridge preservation following extraction. A 76-year-old Caucasian male patient with type 2 diabetes mellitus (HbA1c: 6.2%) presented to the Graduate Periodontics clinic with a fractured crown on #8 that was diagnosed as a non-restorable tooth. The remaining root was extracted. The site was thoroughly debrided and irrigated with saline. The extraction site was grafted using cortical mineralized freeze-dried bone allograft (FDBA). A CO2 laser was used over the bone graft with 1.0-watt continuous irradiation and continuous oscillation setting (coagulation mode) to stabilize the blood clot over the extraction site. No sutures or membranes were used for this procedure. The patient stated he did not need to take any pain medication post surgically, nor feel any FDBA particles come out from the socket. Almost complete closure of the soft tissue was evident at 3 weeks. Implant placement was completed 11 weeks following extraction and socket grafting. Upon implant surgery, adequate bone formation was observed allowing for primary implant stability of over 35 N/cm with an ISQ value of 83. This primary stability enabled the loading of an immediate provisional restoration at time of implant placement. The use of a CO2 laser during implant site development appeared to enhance hard and soft tissue healing which attributed to early implant placement with adequate stability and reduced treatment time for the patient.

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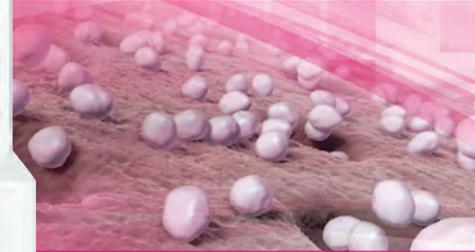


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