

Poster Presentations

All posters will be available for viewing from 2:30 p.m. to 4:30 p.m.

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

2:30 p.m. to 3:30 p.m. Odd-numbered Posters and Clinical Cases

3:30 p.m. to 4:30 p.m. Even-numbered Posters and Clinical Cases

AESTHETIC DENTISTRY

- P1 Effect of Whitening Systems on Stained Enamel and Composites Resins.** S. AL-ANGARI^{1*}, A. SABRAH¹, G. ECKERT², A.T. HARA¹ (¹Indiana University School of Dentistry, ²Indiana University School of Medicine)

The aim of this in-vitro study was to compare the effects of three different bleaching systems on the color stability, roughness and hardness of different stained substrates, including two composite resins (TPH Spectra, Herculite Ultra) and enamel. Box-shaped cavities (3x3x2mm) were prepared in bovine enamel specimens (8x8mm) and restored with the composites. The baseline color, roughness and hardness values were taken for all substrates. Specimens were stained with coffee for two weeks and had the color re-measured. These values were used to randomize specimens into three subgroups: bleaching gel, whitening strips, toothbrushing with whitening toothpaste. Specimens were submitted to a 10-day cycling consisting of treatments with the assigned whitening system and 3-h daily exposure to coffee. At the end of the cycling, color change, roughness and hardness were measured. Data were statistically analyzed using ANOVA at 5%. Gels significantly improve the color compared to other types of whitening in the three substrates ($p < 0.0001$); strips had higher whitening effect compared to toothbrushing in enamel ($p < 0.00001$), while no difference between them was seen for the two composites. Significant higher whitening effect was observed for enamel compared to the two composites, when treated with gel and strips ($p < 0.00001$); with no differences between the composites. However, no difference was observed among these substrates when treated with toothbrushing. There was a significant increase in the roughness for all substrates when treated with the gel, compared to the strips ($p < 0.0364$). None of these treatments differed from toothbrushing. For hardness measurements, no significant differences were found in the hardness changes between the treatments or substrates. In conclusion, gel showed the highest significant improvement in color in the three substrates; however it also increased their surface roughness. Furthermore, enamel was more susceptible to whitening than the two composites.

CARIOLOGY

- P2 Orange Fluorescence in QLF Images of Progressive Carious Lesions.** G.F. GOMEZ^{1*}, G. ECKERT², A.F. ZANDONA³ (¹Indiana University School of Dentistry, ²Indiana University School of Medicine, ³University of North Carolina at Chapel Hill School of Dentistry)

Objective: To determine whether orange fluorescence visually observed in Quantitative Light-Induced Fluorescence (QLF, Inspektor Research System., Amsterdam, Netherlands) images are associated with carious lesions that progressed to cavitation as compared to non-progressive lesions as found by clinical examination. Methods: A retrospective study was conducted on QLF images obtained from 565 children aged 5 to 13 years from Puerto Rico (Ferreira Zandona et al, 2012). Images comprised of occlusal,

buccal and upper lingual surfaces of permanent molars recorded at baseline, 8, 12, 20, 24, 28, 32, 36, 40, 44 and 48 months. There were 1028 surfaces comprising of 267 surfaces that progressed to cavitation based on visual examination using the International Caries Detection and Assessment System (ICDAS) criteria (ICDAS 0/1/2/3/4 to 5/6 or filling), and 761 surfaces that did not progress. Orange fluorescence was determined visually (Y/N) by a single examiner and 360 images were repeated to determine intra-examiner repeatability. Statistical analysis was based on ICDAS scores, different tooth surfaces and visits. Generalized linear mixed-effects models were used to determine the association of orange fluorescence with lesion progression adjusted for baseline ICDAS score, tooth surface and visit. Results: There was no significant association between the presence of orange fluorescence and progression of carious lesion surfaces ($p>0.05$). Orange fluorescence was observed in 91% of the images at least once in the follow-up visits. No significant interactions were found between orange fluorescence and ICDAS baseline score, visit or surface. There was no association of orange fluorescence and progression of caries ($p=0.34$). Intra-examiner repeatability had a 0.70 kappa. Conclusion: Ubiquitous presence of orange fluorescence on most images contributed to the current findings of no significant association between fluorescence and caries progression. Future quantitative analysis of the images with the RF analysis software in QLF-D Bi luminator QA2 version 1.23 is indicated to validate the results. (Supported by NIDCR grant R01 DE017890-05)

P3 Susceptibility of Partially-Desalivated Rats to Dental Erosion by Calcium-Supplemented Beverages. M.A. ALDOSARI^{1*}, T. SCARAMUCCI¹, S.S. LIU¹, J.M. WARRICK-POLAKOFF¹, D. ZERO¹, G.J. ECKERT², A.T. HARA¹ (¹Indiana University School of Dentistry, ²Indiana University School of Medicine)

Objectives: To investigate: 1. the susceptibility of partially desalivated rats to dental erosion; 2. the anti-erosive effect of a calcium-supplemented beverage; 3. micro-computed tomography method for dental erosion quantification. Methods: Thirty-eight male, 3-month old, inbred Fischer 344 rats were used. Half ($n=19$) underwent surgical partial-desalivation by removing the sublingual and submandibular salivary glands. After recovery, they were randomly subdivided into 3 groups ($n=6-7$): A. Sprite Zero (pH 3.35), B. Sprite Zero+20mmol calcium lactate (pH 4.04), C. Deionized water (negative control). Dental erosion simulation was performed for 28 days, where the animals were provided with the drinking fluids and standard rodent laboratory food ad-libitum. The rats were euthanized and their mandibles dissected and fixed with 10% phosphate-buffered formaldehyde. Each left hemi-mandible was coated with paraffin and scanned using micro-computed tomography (Skyscan 1172), at resolution of $8.12\mu\text{m}/\text{pixel}$. Raw image data was reconstructed and the 3 molars used to calculate total enamel volume (mm^3). Pictures of the hemi-mandibles were taken for visual analysis of severity (from 0: no erosion to 3: advanced erosion), by 3 independent and blind calibrated examiners. Data were analyzed by two-way ANOVA and Mantel-Haenszel tests ($\alpha=0.05$), respectively. Results: Microcomputed tomography revealed no significant differences between partially- and non-desalivated groups. Rats consuming A had lower enamel volumes than those consuming B ($p=0.0002$) or C ($p=0.0002$), which did not differ from each other ($p=0.88$). For visual analysis, desalivation did not affect erosion ($p>0.05$). Rats consuming C showed the lowest erosion scores, followed by B and then A (all $p<0.05$), for both partially and non-desalivated. The Spearman correlation between the two outcomes was -0.65 ($p<0.001$). Conclusions: 1. Partially-desalivated rats did not show more dental erosion; 2. Ca-containing beverage was effective preventing dental erosion; 3. Micro-tomography was able to quantify dental erosion, although it was not as sensitive as the visual method.

P4 Relative Fluoride Response of Caries Lesions Created in Fluorotic and Sound Teeth Studied Under Remineralizing Conditions. H. ALHAWIJ*, F. LIPPERT, E. MARTINEZ MIER (Indiana University School of Dentistry)

Objective: The present in vitro pH cycling study investigated potential differences between caries lesions created in fluorosed and sound enamel with regards to their responsiveness to fluoride under remineralizing conditions. Methods: Human molars (sound and fluorosed) were divided into four groups based on their Thylstrup-Fejerskov score (TF0-3). Each group was further divided into two treatment groups (n=45 each): deionized water or 383 ppm fluoride. Artificial enamel caries lesions were created and pH cycled for 20 days using an established net remineralization model. Quantitative light-induced fluorescence was used throughout the study to investigate lesion severity and changes thereof. Data were analyzed using two-way ANOVA. Results: There were no differences in lesion severity between all groups after lesion creation ($p_{lesion}=0.1934$). The TF score vs. treatment interaction was significant at all other time points ($p_{10d}=0.0280$; $p_{20d}<0.0001$; $p_{secdem}=0.0411$). Relative differences in responsiveness to fluoride vs. deionized water increased with increasing TF scores. In comparison to lesions created in sound enamel, lesions created in enamel with moderate fluorosis (TF 2/3) were more prone to remineralization in the presence than in the absence of fluoride. Furthermore, lesions created in enamel with moderate fluorosis exhibited more remineralization in the presence of fluoride than lesions created in sound teeth, whereas the opposite was true for deionized water. Conclusion: The extent of enamel fluorosis severity directly impacts subsequent lesion re- and progression as well as the lesion's responsiveness to fluoride. Clinical relevance: The results of this study will facilitate the understanding of the beneficial effects of fluoride on fluorosed teeth.

DENTAL MATERIALS

P5 Effect of Chitosan Coating on BSA Adsorption on Bioactive Glasses. B. BALLENGER*, W. CHING LIU, T.G. CHU (Indiana University School of Dentistry)

Objective: This study investigated the protein release kinetics from bioactive glasses with or without boron-doping (13-93B3 and 13-93B0) and evaluated the effects of chitosan coatings on the release and secondary structures of protein on 13-93B0. Methods: 13-93B0 and 13-93B3 BG discs were fabricated and sintered at 650 °C (B0) and 540 °C (B3). For BSA release assay, 4-mg BSA was loaded onto sample discs (diameter=13-mm, thickness=1.9-mm) coated with 80% or 94% deacetylated (DEA) chitosan at concentrations 0.5, 2 and 4% for one time or 2% for three times. Discs were immersed in 2-mL PBS which was collected and replenished at time points between 1-168 hours. BSA release was evaluated using BCA Protein Assay. Degradation study was performed on B0 and B3. Secondary structures of BSA were characterized using FTIR. Results: 100% protein release occurred by 24-hours for all B3 groups. Chitosan coating delayed 100% release from B0 2-80 and B0 2-94 to 72-hours. Different chitosan coatings on B0 yielded 100% release at 24-hours for B0-UC & B0-2%, at 72-hours for B0-2% SW and 168-hours for B0-3x2%. For FTIR analysis, the highest α -helix content was found in the 2% sandwich chitosan coating group of the value of $31.9 \pm 2.1\%$, which was significantly different from uncoated and 4% chitosan coating groups. Conclusion: The faster BSA release in B3 groups matches with the faster degradation properties of B3; however, chitosan coating could effectively delay BSA release from B0 glass. Sandwich coating was studied because glass and BSA are negatively charged while chitosan is positively charged. The α -helix content suggests the better prevention of BSA unfolding of the sandwich coating on B0 glass. In conclusion, chitosan coating is demonstrated to delay the release and restore the conformation of BSA on B0 glass.

P6 Effect of Intracanal-medicaments and EDTA on Indentation Properties of Dentin. G. YASSEN* and J.A. PLATT (Indiana University School of Dentistry)

Objective: The aim of this study was to investigate the effect of intracanal antibiotic medicaments used in endodontic regeneration and ethylenediaminetetraacetic acid (EDTA) on the microindentation properties of root canal dentin surfaces using a reference point indentation technique (RPI). Methods: Immature human premolars were selected (n =10). Four specimens were obtained from each root without touching the root canal surface and randomly assigned to three treatment groups and a control group. Each specimen was exposed to either double antibiotic paste (DAP) for four weeks followed by EDTA for five minutes, triple antibiotic paste (TAP) for four weeks followed by EDTA for five minutes or Hank's balanced salt solution for four weeks followed by EDTA for 5 minutes or Hank's balanced salt solution for four weeks (control). The indentation properties of the dentin surfaces were measured using a BioDent reference point indenter according to the following protocol: indentation force (N) = 5; indentation speed (Hz) = 2; indentations per measurement (Cycles) = 10. One-way ANOVA and Fisher's protected least significant differences were used for statistical analyses ($\alpha=0.05$). Results: The three treatment groups caused significant increase in first-cycle indentation distance (ID) and total indentation distance (TID) compared to the untreated control group ($p<0.0001$). Microhardness values were significantly lower in the three treatment groups compared to the untreated control group ($p<0.0001$, $p=0.0021$). Furthermore, dentin groups treated with antibiotic paste followed by EDTA had significantly lower microhardness than EDTA treated dentin ($p<0.0001$). Conclusion: Significant differences were observed in the majority of indentation parameters of the root canal surfaces treated with various endodontic regeneration medicaments and/or EDTA compared to an untreated control. BioDent RPI may offer an advantage of mechanical characterization of root canal predentin in immature teeth without any metallographic preparation.

P7 Antibacterial Effect of Nanofibrous Metronidazole-Containing Drug-Delivery Systems for Periodontal Applications. A.C. STARR*, K. KAMOOCKI, R.L. GREGORY, M.C. BOTTINO (Indiana University School of Dentistry)

Periodontitis is a chronic inflammatory disorder that affects the integrity of the periodontium. Though antimicrobials have been systemically administered as an adjunct to the surgical removal of pathogens its overuse has been associated with side effects such as bacterial strain resistance. The present study was designed to evaluate the antimicrobial efficacy of nanofibrous metronidazole-containing drug-delivery systems against *P. gingivalis* (*Pg*) over time. A biodegradable polymer blend based on poly(DL-lactide-co-glycolide; PLGA) and type-B gelatin (PLGA:GEL) was prepared in hexafluoroisopropanol. Metronidazole (MET) at 5 and 25wt.% concentrations were added to the blend and electrospun into nanofibers. PLGA:GEL samples were processed to serve as control. Electrospun samples (n=3) were sized based on a predetermined weight ($4.0\pm 0.2\text{mg}$), sterilized and incubated in 2mL of sterile PBS. Samples were maintained in an incubator at 37°C up to 14 days. 500 μL aliquots were taken from the incubation medium at 1, 3, 7 and 14 days. Equal amounts of fresh PBS were added following aliquots collection. *Pg* was cultured anaerobically for 48h (37°C) in BHI broth containing 5g/L yeast extract with 5% v/v Vitamin K/hemin. Bacteria were spread onto blood agar plates and then 15 μL of the retrieved aliquots pipetted in triplicate. Sterile water and 0.12% chlorhexidine solution served as controls. Samples were incubated anaerobically for 48h and inhibition zones were measured. Data were analyzed at 5% significance level. All aliquots collected from the incubated MET-containing nanofibrous drug-delivery

systems inhibited the growth of *Pg*. The highest inhibition was observed at Day 1, with a sustained bacterial growth inhibition over time. As expected 25wt.% MET aliquots provided significantly ($p < 0.05$) greater inhibition compared to the 5wt.%, for all time points except Day 7. MET-containing nanofibrous drug-delivery systems provided a sustained antimicrobial response over 2 weeks. Drug-release kinetics and cytocompatibility with periodontal-related cells are ongoing.

P8 Effects of Bi-Mix Antimicrobial Scaffolds on Human Dental Pulp Stem Cells. K. KAMOCKI* and M.C. BOTTINO (Indiana University School of Dentistry)

Introduction: Antibiotic pastes used for bacterial disinfection prior to regenerative endodontic procedures have demonstrated to promote major adverse effects on dental stem cells survival and viability. Meanwhile, recently developed antibiotic-containing nanofibrous scaffolds have shown not only substantial antimicrobial properties but also significant promise towards a more cell-friendly alternative. Here we report the indirect effects of these antibiotic-containing scaffolds on human dental pulp stem cells (hDPSCs) viability and proliferation. Methods: hDPSCs were exposed to scaffolds containing both metronidazole and ciprofloxacin at selected weight ratios. The following groups of polydioxanone-based (PDS) scaffolds were tested, namely: pure metronidazole/MET, pure ciprofloxacin/CIP, 3:1MET/CIP, 1:1MET/CIP; and 1:3MET/CIP. Antibiotic content was 25wt.% of the total scaffold weight (i.e., 150 mg of drug alone or combined). Pure PDS scaffolds served as negative control. Saturated solution of CIP/MET in complete culture medium was prepared to mimic the double antibiotic paste (DAP) and served as a positive control. WST-1 assay was used to evaluate the impact of the distinct scaffolds on hDPSCs proliferation after 3, 5, and 7 days. LIVE/DEAD® assay was used to determine viability of hDPSCs after 3 days of exposure. High performance liquid chromatography (HPLC) was conducted to investigate the amount of drug(s) released from scaffolds over 2 weeks. Results: Exposure of hDPSCs to all CIP-containing scaffolds revealed reduced proliferation ($p < 0.05$) and viability, with a concentration dependent trend, whereas no changes were observed in groups exposed to scaffolds with MET only. Conclusions: The use of electrospun biodegradable antibiotic-containing scaffolds as a delivery system to promote root canal disinfection prior to the regenerative endodontics procedure may have less detrimental effect on pulp stem cells viability and proliferation, thus facilitating dental pulp regeneration. (This work was supported in part by a Grant from the American Association of Endodontists Foundation and start-up funds from the IU School of Dentistry (M.C.B.))

P9 Chemical and Mechanical Characterization of a Novel Urethane-Based Composite. A. ALZAIN^{1*}, J.A. PLATT¹, D. XIE² (¹Indiana University School of Dentistry, ²Department of Biomedical Engineering, Purdue School of Engineering and Technology, IUPUI)

The aim of this study was to develop a novel urethane-based resin composite with comparable degree of conversion (DC) and flexural strength (FS) to traditional matrix resin composites. The DC was measured using attenuated total reflection Fourier transform infrared (ATR-FTIR) spectroscopy ($n=3$) at different curing times (20, 40 and 60 seconds). Furthermore, FS was performed according to ISO 4049 ($n=5$) using a universal mechanical testing machine (MTS) at a crosshead speed of 1 mm/min. Both FS and DC of the two novel urethane-based resin composites (M1 and M2) were evaluated at three different weight ratios (25/75, 55/25 and 75/25) mixed with a diluent (Triethylene glycol dimethacrylate-TEGDMA). The novel materials were compared with two resin-based composite controls (urethane dimethacrylate-UDMA and Bisphenol A glycidyl methacrylate-Bis-GMA) at 55/45-weight ratio. Data was analyzed using ANOVA followed by Tukey tests ($\alpha=0.05$). The DC was not significantly different between curing times for each resin composite ($p=0.079$) at 55/45 ratio. However, DC of M2/TEGDMA was significantly lower compared to the other groups ($p < 0.001$). There was no significant interaction between resin composite materials and curing times ($p=0.150$). For FS, the 55/45 and 75/25 ratios of novel resin composites (M1/TEGDMA

and M2/TEGDMA) met the ISO 4049 FS criteria. M1/TEGDMA at 55/45 ratio showed significantly higher elastic modulus compared to UDMA/TEGDMA. Additionally, FS of M1/TEGDMA at 55/45 ratio was not significantly different from the controls. We conclude that M1/TEGDMA novel urethane-based resin composite at 55/45 ratio has comparable DC and FS to traditional matrix resin composites. Further studies are needed to investigate the potential ability of M1/TEGDMA to resist degradation.

P10 Attitudes of Dental Hygiene Students on Rubber Dam Isolation. D. EL-HOUT*, J. JAEGER, S. CHITRE (University of Detroit Mercy School of Dentistry)

Objective: The aim of this study is to determine disposition on rubber dam isolation (RDI) technique. Dentists have utilized a rubber dam to isolate an operative field. Isolation is a critical step and should be used whenever possible according to “The Quality Assurance Guidelines of the American Associations of Endodontists”. It is key to use RDI when delivering endodontic therapy to avoid complications including ingestion of files or toxic liquids. A contraindication for use of RDI may include an allergic reaction to the rubber dam material. Another contraindication is if the tooth shows gross decay in which an instrument such as forceps cannot be utilized properly with a rubber dam in place. Methods: Investigators conducted a survey of the class of 2013 and 2014 dental hygiene students regarding their attitudes towards using and placing RDI. This survey was approved by the UDM Institutional Review Board (Protocol #1213-10). Results: There were 33 participants. 96% of the participants use RDI for isolation purposes, while 40% stated they would use it to prevent lawsuits and for better access. 91% stated that a disadvantage of the RDI is that it is time consuming. A majority of the students stated that they have experienced a clinical situation where placing RDI was difficult. 76% of the students stated that they believe the utilization of RDI is important. Conclusion: 68% of the participants would use other isolation methods besides RDI after graduating dental hygiene school. The reason for that is because it is time consuming. Also, RDI would not always work for the clinical situation at hand. In many instances, cotton roll isolation or a dry angle would be used instead of Rubber Dam Isolation.

P11 Optimization of Crystalline Kinetics, Thermal Processing, and Strength of a Dental Lithium Disilicate Glass-ceramic. W. LIEN^{1,3*}, H. W. ROBERTS², T.G. CHU³ (¹Air Force Institute of Technology, USAF, ²Keesler Air Force Base, USAF, ³Indiana University School of Dentistry)

Background: Elucidating the lithium disilicate system like the popular IPS e.max® CAD (LS2), made specifically for Computer-Aided Design and Computer-Aided Manufacturing (CAD-CAM), as a function of temperature unravels new ways to enhance material properties and performance. Objective: To study the effect of various thermal processing on the crystallization kinetics, crystallite microstructure, and strength of LS2. Methods: The control group of the LS2 samples was heated using the standard manufacturer heating-schedule. Two experimental groups were tested: (1) an extended temperature range (750-840 °C vs. 820-840 °C) at the segment of 30 °C/min heating rate, and (2) a protracted holding time (14 min vs. 7 min) at the isothermal temperature of 840 °C. Five other groups of different heating schedules with lower-targeted temperatures were evaluated to investigate the microstructural changes. For each group, the crystalline phases and morphologies were measured by X-ray diffraction (XRD) and scanning electron microscope (SEM) respectively. Differential scanning calorimeter (DSC) was used to determine the activation energy of LS2 under non-isothermal conditions. A MTS universal testing machine was used to measure 3-point flexural strength and fracture toughness, and elastic modulus and hardness were measured by the MTS Nanoindenter® XP. A one-way ANOVA/Tukey was performed per property (alpha = 0.05). Results: DSC, XRD, and SEM revealed three distinct microstructures during LS2 crystallization. Significant differences were found between the control group, the two aforementioned experimental groups, and the five lower-targeted-temperature groups per property (p<0.05). The activation energy for

lithium disilicate growth was 667.45 (\pm 28.97) KJ/mole. Conclusions: Groups with the extended temperature range (750-840 °C) and protracted holding time (820-840 °C H14) produced significantly higher elastic-modulus and hardness properties than the control group but showed similar significant flexural-strength and fracture-toughness properties with the control group. In general, explosive growth of lithium disilicates occurred only when maximum formation of lithium metasilicates had ended.

ENDODONTICS

P12 Comparison of Propolis Antimicrobial Activity Using Three Delivery Methods. M. DAETWYLER*, Y. EHRLICH, R.L. GREGORY (Indiana University School of Dentistry)

Propolis is a non-toxic product of honeybees, which contains flavonoids, phenols and aromatic compounds. Ecuadorian Rainforest, LLC (EP) is propolis product that has been shown to be beneficial in the treatment of a longstanding endodontic infection. *In vitro* studies at IUSD have shown that EP has antibacterial properties against *Fusobacterium nucleatum* prominent in primary endodontic infections. Different solvents are used to dissolve propolis and the propolis extracts can be used as intracanal medicaments. The purpose of this *in vitro* study is to compare the antimicrobial efficacy of EP extracts from three different solvents. *Enterococcus faecalis* and *Candida albicans* were chosen as the test organisms. *E. faecalis* is mostly found in secondary endodontic infections and *C. albicans* plays a pivotal role in biofilm formation. The solvents used were dimethyl sulfoxide (DMSO), propylene glycol, and ethanol. EP was dissolved in each solvent, the solution was centrifuged, and the supernatant was taken to test for antimicrobial activities. *E. faecalis* and *C. albicans* were then grown in broth media with increasing concentrations of the propolis extract. After incubating for 48 hours in 96 well plates, total growth was measured using a SpectraMax 190 spectrophotometer. 120 μ l aliquots were separated to measure planktonic growth. Finally, by fixing the biofilm to the wells and staining with crystal violet the biofilm formation could be measured. The results show clear inhibition of total growth in all three propolis containing solvents in a dose-dependent manner. However, further testing revealed strong antimicrobial activity in the higher concentrations of solvent alone. This perhaps masks the effect of propolis and does not enable us to evaluate the effect of the propolis independent of the solvent used. Further research would need to allow high concentrations of propolis without the high concentration of solvent.

P13 Clindamycin-Containing Scaffolds for Root Canal Disinfection. M.M. SLY*, K. KAMOCKI, R.L. GREGORY, K.J. SPOLNIK, and M.C. BOTTINO (Indiana University School of Dentistry)

Clindamycin seems to be ideally useful for applications in the endodontic system, in addition to being widely employed by endodontists, though systemically. Clindamycin has strong antimicrobial activity against many oral pathogens. One of its adverse effects is the Clostridium difficile-associated diarrhea. If Clindamycin is not used systemically, but confined into the root canal system, the risk associated with this adverse effect would possibly be eliminated. The aim of this study was to fabricate/characterize Clindamycin-containing scaffolds and investigate their antimicrobial effects against *C. albicans*, *E. faecalis*, and *S. mutans*. Polydioxanone/PDS was dissolved (10 wt.%) in hexafluoropropanol. Clindamycin (Sigma) was added and mixed with the PDS solution at 5%, 15%, and 30 wt.%. Pure PDS and clindamycin-containing PDS solutions were electrospun into scaffolds under optimized parameters. Scanning electron microscopy/SEM was used to analyze fiber diameter/distribution. Tensile strength of the scaffolds was evaluated under dry and hydrated conditions. Antimicrobial efficacy, via agar diffusion assay, was determined against *C. albicans*, *E. faecalis*, and *S. mutans*. 5-mm in diameter scaffold samples were disinfected with UV light, and placed in blood agar plates containing *Ca*, *Ef*, and *Sm*. After 3 days of incubation, the inhibition zones were determined (mm). Clindamycin incorporation led to a significant ($p < 0.05$) reduction in fiber diameter (5%=264 \pm 73nm, 15%=282 \pm 68nm, and 30%=332 \pm 89nm

when compared to the control (PDS=1160±404nm). The tensile strength values of hydrated 15 and 30% clindamycin-containing scaffolds were significantly ($p<0.05$) higher than those of all other groups. Clindamycin-containing samples inhibited growth of all pathogens tested. The largest inhibition zones were seen on *Ca*. Conclusions: Clindamycin-containing scaffolds were successfully synthesized through electrospinning. Based on the data collected, clindamycin-containing scaffolds show promise towards applications in endodontics as a disinfection strategy prior to regenerative endodontic procedures. (This work was supported in part by a Grant from the American Association of Endodontists Foundation and start-up funds from the IU School of Dentistry (M.C.B.))

MICROBIOLOGY/IMMUNOLOGY/ORAL BIOLOGY

P14 Toxicity and Cytokine Release Effects of Fulvic Acids on Human Gingival Fibroblasts. B. ALAVANJA*, L.J. WINDSOR (Indiana University School of Dentistry)

Background: Periodontitis is a chronic disease characterized by inflammation that leads to the destruction of tooth supporting tissues. Although the primary etiology of periodontitis is bacterial plaque, it is now understood that host responses are the main cause of tissue breakdown. Cells, including human gingival fibroblasts (HGFs), release inflammatory cytokines that lead to periodontal destruction. Fulvic acids are naturally occurring organic compounds classified as humic substances. Investigators found that fulvic acids produce anti-inflammatory effects. Fulvic acids may be a natural alternative to conventional anti-inflammatory medications and may be used for modulation of host response with respect to periodontitis. Purpose: The purpose of this *in vitro* study was to examine the toxicity of specific fulvic acids with respect to HGFs and evaluate these fulvic acids effects on cytokine release from unstimulated HGFs and HGFs that have been stimulated with *Porphyromonas gingivalis* supernatant. Methods: HGFs already cultured from samples of clinically non-inflamed gingival connective tissues were used with Institutional Review Board approval. HGFs were incubated with fulvic acids (concentrations 0-4%) to determine cell cytotoxicity. HGFs stimulated and unstimulated with 10% *P. gingivalis* supernatant were then incubated with or without 0.5 % fulvic acids for cytokine release analyses using a cytokine micro array technique. Results: Cytotoxicity assays determined 0.5% to be the highest concentration of fulvic acids that were nontoxic to HGFs. Cytokine array analysis showed reduction in IL-6 production for unstimulated fulvic acids treated HGFs compared to untreated controls. Also, IL-8 levels were reduced in stimulated fulvic acids treated HGFs compared to untreated controls. Conclusion: Fulvic acids treatment alters the cytokine production profiles of both *P. gingivalis* stimulated and unstimulated HGFs *in vitro*, reducing pro-inflammatory cytokine production.

P15 Study of Fulvic Acid: A Natural Dietary Supplement. G.I. SYED^{1*}, R.L. GREGORY², L.J. WINDSOR², F. SONG² (¹Department of Biology, Indiana University Purdue University Indianapolis, School of Science, ²Department of Oral Biology, Indiana University School of Dentistry)

Shilajit is a substance found in parts of Asia. Although there have been no clinical studies, it is used by the locals as an antiseptic, anti-inflammatory and pain suppressor. Fulvic acid (F-A) is a major constituent of shilajit and was used to test the anti-pathogenic tendencies of shilajit on bacteria and the cytotoxic effects on human cells in the oral cavity. The bacterial study was performed on *Streptococcus mutans*, a normal flora of the oral cavity. The idea was to test the metabolic activity of the bacteria in F-A-containing media. Menadione-XTT reagent was used for this. The bacterial biofilm was allowed to grow in TSBS in a microtiter plate of 96 wells. The F-A solution of different concentration were introduced into each well in a gradually decreasing amount and the last control wells had a zero concentration. The XTT reagent was introduced and after incubation the biofilm of *S. mutans* reduced the XTT to an orange color depending

on its overall metabolic activity, the change in color was detected by measuring the absorbance at 490nm. Between 2.5-5.0% (v/v) of F-A the wells showed signs of decreased activity. The cell proliferation and cytotoxicity of F- A on human pulp and pulp stem was tested utilizing WST and LDH assay, respectively. The results from the bacterial study indicated that absorbance of the wells with concentrated F-A was lower compared to the wells with more diluted F-A solutions. These results demonstrated that F-A had a negative effect on the growth and metabolic activity of *S. mutans*. Overall, the experiment validates the potency of F.A as an effective antibacterial. Further testing is needed but the compound shows promise and can be employed as an effective ingredient of mouthwash and other such antiseptic products. (Support by the IUPUI Center for Research and Learning)

P16 Direct and Indirect Cytotoxicity of Antibiotics Medicaments for Endodontic Regeneration.
A.H. SABRAH^{1*}, W.S. GOEBEL², R.L. GREGORY¹, J.A. PLATT¹ (¹Indiana University School of Dentistry, ²Indiana University School of Medicine)

The reported cytotoxicity to stem cells associated with the use of antibiotic pastes (1000 mg/mL) raise concerns about their suitability as intracanal medicaments for endodontic regeneration. The aim of this study is to identify the antibiotic concentrations that can provide a direct (after application) and indirect (Substantive properties after removal) optimized antibacterial action without compromising the survival of human dental pulp stem cells (hDPSC). The direct cytotoxic effect of antibiotic dilutions (Triple antibiotic (TAP) and Double antibiotic (DAP) pastes/ 10, 1, 0.5, 0.25, and 0.125 mg/ml) on *Enterococcus faecalis* established biofilm, and the direct cytotoxicity against hDPSC were investigated using spiral plating and colorimetric (WST-1/LDH) cytotoxicity assays, respectively. To measure the indirect antibacterial activity and cytotoxicity, Human dentin specimens (4x4x1 mm³) were immersed in different concentrations of antibiotics (0.5, 1, and 1000 mg/ml) for 14d. Specimens were irrigated with saline and the ability of *Enterococcus faecalis* to grow on the specimens were measured after 0 d, 3 d, 7 d, and 14 d of incubating the specimens in PBS solutions. The cytotoxicity of released antibiotics in the PBS solutions against hDPSC was measured. Each experiment was repeated at least three times with triplicate wells/specimens in each experiment. Mixed linear model with pair-wise comparison was used to analyze the data for each experiment ($\alpha=0.05$). Antibiotic dilutions were able to significantly decrease the bacterial CFU/ml ($p<0.05$). All tested antibiotic dilutions except 0.125 mg/ml decrease the proliferation of hDPSC. DAP dilutions of 1 mg/ml and 0.5 mg/ml were found to have equal indirect protective antibacterial effects comparable to antibiotic pastes over tested time points with no indirect cytotoxicity to hDPSC. The use of diluted antibiotic medicaments can provide a significantly direct antibacterial action, significantly higher survival of hDPSC, and a significantly comparable indirect protective antibacterial action as compared to the concentrated paste form.

P17 Modification of the Limulus Amoebocyte Lysate Assay for Endotoxin Detection in Canine Blood Specimens. D.R. WAGENKNECHT^{1*}, L.F. GUPTILL², D.M. GALLI¹ (¹Indiana University School of Dentistry, ²Purdue University College of Veterinary Medicine)

Endotoxins (lipopolysaccharides) are a major component of the cell wall of gram-negative bacteria. It is proposed that translocation of endotoxin from the oral cavity into the bloodstream increases the risk for systemic disease in periodontitis patients. Indeed, periodontal therapy results in reduction of endotoxin levels leading to improvement of systemic inflammation. An association of canine periodontal disease with renal and cardiovascular disease has been suggested, but research is needed to determine if endotoxemia and chronic low-grade inflammation are pathogenic components of canine periodontal disease. Objective: This study was designed to determine levels of endotoxin in the blood of greyhounds (n=12) before and after undergoing routine dental care. Methods: Serum and heparin plasma were collected immediately prior to cleaning, and 4 and 12 wks post-cleaning. A bioassay, Limulus

Amoebocyte Lysate (LAL), was selected for measurement of endotoxin units (EU). Since blood contains inhibitors of LAL, sample treatment and protocol modifications were examined. The effects of heat treatment, dilution factor, Pyrospense™ and MgCl₂; and the optimal diluent for the standard curve were examined. Each sample was spiked with 0.05 EU/ml of *Escherichia coli* endotoxin standard to compare % recovery of endotoxin in serum and plasma. All samples were duplicate tested in three independent LAL assays; mean EU values were evaluated. Results: Endotoxin recovery was higher in serum than plasma. When compared to baseline levels, serum collected at 4 and 12 weeks showed increased endotoxin for 33% and 50% of the dogs respectively. Endotoxin levels did not decrease post-treatment as anticipated. Conclusion: LAL assay variability may be a confounder of the data in that standard deviations of replicate testing were high. Additional investigations are required to understand the dynamics of canine serum in the LAL assay. (Supported by grants from the IUPUI Office for Research and the Purdue University College of Veterinary Medicine)

P18 Porphyromonas gingivalis Biofilm Growth Inhibition by Arginine Deiminase M.B. KANE*, K.S. GREGSON, G.G. ANDERSON, R.L. GREGORY (Indiana University School of Dentistry)

Porphyromonas gingivalis is found in approximately 80% of patients who have periodontal disease. The ability of this species to form a dental biofilm strengthens its virulence. *Streptococcus cristatus* has been shown to inhibit the ability of *Pseudomonas aeruginosa* to form this biofilm. It is believed that arginine deiminase (ADI) in the *S. cristatus* membrane is responsible for this inhibition. The aim of this research was to investigate the effect of isolated ADI on the growth of *P. gingivalis* biofilm. *P. gingivalis* cultures treated with varying concentrations of *S. cristatus* whole cell lysate with and without membrane ADI, *S. cristatus* membranes with and without membrane ADI, and isolated ADI were incubated for 48 hours in BHI + yeast extract + vit K + hemin. After treatment with formaldehyde, crystal violet, and isopropanol, absorbance was measured to determine growth of biofilm. The isolated ADI assay demonstrated significant ($p < 0.05$) promotion of *P. gingivalis* biofilm growth at a concentration of 0.500 μ M and significant ($p < 0.05$) inhibition of *P. gingivalis* biofilm growth at concentrations of 0.950 μ M and up. There was no significant ($p > 0.05$) difference between *S. cristatus* whole cell lysates with or without the ADI protein in the membrane. There was no significant ($p > 0.05$) difference between *S. cristatus* membranes with or without the ADI protein in the membrane. The results suggest that while low concentrations of ADI promoted growth of the biofilm, higher concentrations rapidly inhibited growth. Despite *P. gingivalis* biofilm growth being inhibited when grown in conjunction with *S. cristatus*, wild type *S. cristatus* whole cell lysate and membranes did not show a statistical difference in their effect on biofilm growth.

P19 The Effect of Fulvic Acid on Streptococcus mutans and SCC-25 Cells. P. WITCHER*, R.L. GREGORY, L.J. WINDSOR (Purdue School of Science, IUPUI, Indiana University School of Dentistry)

Shilajit is a homeopathic treatment used by local inhabitants of India and Pakistan. It may have specific components that inhibit the formation of cavities and the growth of cancer cells. This experiment analyzed the effects of fulvic acid, an active component of shilajit, on the growth of oral bacteria and squamous cell carcinoma. The effect of fulvic acid was evaluated on early *Streptococcus mutans* (*S. mutans*) biofilm formation and established *S. mutans* biofilm by treating each group with different concentrations of fulvic acid for 24 hours in sterile 96-well flat-bottom microtiter plates. *S. mutans* was used because it is a common cause of dental caries. The optical density (OD) of the *S. mutans* biofilm was measured after crystal violet staining using a SpectraMax190; greater growth correlated to greater OD. It was determined that fulvic acid inhibits the growth of newly forming *S. mutans* biofilm at fulvic acid concentrations greater than 1.25% (vol. %) and established *S. mutans* biofilm at fulvic acid concentrations greater than 5% (vol. %). To evaluate the effect of fulvic acid on squamous cell carcinoma (SCC-25) cells, six-well plates

seeded with SCC-25 cells (1×10^5 cells/well) were exposed to different concentrations of fulvic acid (buffered to a pH of 7.5) for 72 hours. The cytotoxicity and cell proliferation were measured using a cytotoxicity detection kit and a water soluble tetrazolium kit (Roche Applied Science), respectively. It was determined that fulvic acid inhibits the growth of SCC-25 cells at concentrations of fulvic acid above 2% (volume %). Fulvic acid (0.50% by volume) also inhibited the ability of SCC-25 to degrade collagen compared to the control. The suppressive mechanisms observed by fulvic acid on both *S. mutans* and SCC-25 cells could improve overall oral health through a mouthwash application.

P20 Antimicrobial Effects of Magadi Trona on *Streptococcus mutans* Biofilms. L.L. ALBRECHT*, K.M. YODER, R.L. GREGORY (Indiana University School of Dentistry)

Magadi, $\text{Na}_2\text{CO}_3 \cdot \text{NaHCO}_3 \cdot 2\text{H}_2\text{O}$, is a sodium rich mineral which also contains fluoride, that is found in salt deposits surrounding alkaline lakes and areas on surface soil that experiences high evaporation rates in Kenya and Tanzania. This trona product is a cooking additive used for tenderization purposes, to increase cooking efficiency, and as a flavoring agent in the villages of Kenya and Tanzania. The purpose of this study was to determine if magadi had an antimicrobial effect on oral biofilm formation of *Streptococcus mutans*. Objective: *S. mutans* biofilms, with and without magadi, were established in the laboratory, to reproduce biofilms that are present in the oral environment. In studies examining fluoride in relation to caries prevention, there is evidence that fluoride-containing products have antimicrobial effects. There are more than 700 bacterial species found in the oral microflora, but the primary bacterium involved in caries development, and the transmission of caries is *S. mutans*, a Gram-positive facultative anaerobic bacteria. Methods: Cultures of *S. mutans* cells in tryptic soy broth (TSB) were diluted to 1:200 with TSB containing 1% sucrose (TSBS). A prepared concentration of filter-sterilized magadi solution (3g magadi/100ml distilled water) was added. The magadi solution was diluted in TSBS. The dilutions were incubated in sterile 96-well microtiter plates for 24 to 72 hours at 37°C. Results: The assays demonstrated that 30mg/ml magadi completely inhibited the growth ($p < 0.05$) of *S. mutans*. However, the presence of moderate levels of magadi concentrations (0.5-7.5mg/ml) significantly enhanced (up to two fold; $p < 0.05$) biofilm formation. The minimum inhibitory concentration was determined as 15mg/ml. Conclusion: The results obtained provide evidence magadi in high concentrations inhibits oral bacteria involved in caries development. Magadi may contribute to preventing the caries process.

P21 Determining the Minimum Inhibitory Concentration of Fulvic Acid on Early *Streptococcus mutans* Biofilm Formation. A. TAHIR*, P. WITCHER, R.L. GREGORY (IUPUI Purdue School of Science, Indiana University School of Dentistry)

Fulvic acid is an active ingredient in many homeopathic treatments. Shilajit is one such treatment that is mainly used in the Indian Subcontinent area. It is believed that fulvic acid has some remedial properties that can help with inhibition of many types of bacteria and various malignant diseases. In this experiment the effects of fulvic acid were analyzed on early *Streptococcus mutans* biofilm formation. *S. mutans* is an oral bacterium that contributes to the formation of dental caries forming bacterial biofilm on teeth. For the experiment, early *S. mutans* biofilm formation was treated with different concentrations of fulvic acid for 24 hours in sterile 96-well flat-bottom microtiter plates. The optical density (OD) of the *S. mutans* biofilm was then measured using a SpectraMax190, after staining with crystal violet. Data was analyzed on the knowledge that a greater OD is correlated to a greater bacterial biofilm. Results demonstrated that fulvic acid inhibited the growth of early *S. mutans* biofilm formation at fulvic acid concentrations greater than 5% (vol. %). After preliminary studies, different concentrations of fulvic acid closer to the estimated minimum inhibitory concentration (MIC) were applied to the *S. mutans* to find a more precise MIC of the fulvic acid on the biofilm growth. Upon completion of these various studies, fulvic acid was shown to inhibit early *S. mutans* biofilm formation and may show signs of oral health improvement if applied for human use.

P22 Effects of Nicotine and Polyphenols on *Streptococcus mutans* Binding Endothelium. C. LEMINH*, R.L. GREGORY (Indiana University School of Dentistry)

Cardiovascular disease is the leading cause of death in the United States. *Streptococcus mutans* has a role in the pathogenesis of cardiovascular disease, and it can contribute to atherosclerosis by utilizing antigen I/II adhesion to bind to fibronectin found on the surface of endothelial cells. In the presence of nicotine, the expression of several *S. mutans* virulence factors, including I/II, are enhanced. The purpose of this research was to determine if tea polyphenols (EGCG) would reduce the nicotine-induced expression of these surface adhesins to fibronectin. Objective: To investigate binding of *S. mutans* exposed to various concentrations of nicotine and tea polyphenols to fibronectin. *S. mutans* UA159 was incubated in various nicotine dilutions with and without 0.25 mg/ml of EGCG in order to analyze the binding of the bacterial cells to fibronectin. Methods: *S. mutans* UA159 was cultured in tryptic soy broth (TSB), supplemented with different nicotine concentrations (0-32mg/ml) and nicotine plus EGCG dilutions, and biotinylated. Microtiter plates were then coated with fibronectin and the biotinylated bacterial cells added followed by extra-avidin-peroxidase and O-phenylenediamine (OPD). The binding was measured by absorbance using a spectrophotometer at 490 nm. Results: Universally nicotine only dilutions incubated with *S. mutans* demonstrated significantly more ($p < 0.05$) adhesion to fibronectin than nicotine/EGCG dilutions with *S. mutans*. As the concentration of nicotine was reduced, EGCG caused a 1.7 fold average decrease adhesion to fibronectin compared to the control. Conclusion: Nicotine up-regulates the adhesion of *S. mutans* to fibronectin. EGCG inhibited this increased adhesion. Since the presence of EGCG demonstrated a decrease in bacterial adhesion to fibronectin, these results may suggest a therapeutic role for tea in people who have cardiovascular disease and smoke.

P23 Polyphenol Effect On *Streptococcus mutans* ComC- Biofilm. J. EVANS*, R.L. GREGORY, R. HUANG (Indiana University School of Dentistry)

This study investigated the effect that polyphenols have on *Streptococcus mutans* quorum sensing as it relates to biofilm formation. Polyphenols act as not only antioxidants, but also can be bactericidal. Objectives: The effect of polyphenols on biofilms of a quorum sensing mutant of *S. mutans* was investigated. A *S. mutans* ComC- deficient strain was incubated in different concentrations of competence stimulating peptide (CSP) with epigallocatechin-3 gallate (EGCG) and compared to ComC- without EGCG to help understand polyphenol's anti-biofilm effects. Methods: The ComC- mutant was incubated in Tryptic Soy broth supplemented with 1% sucrose (TSBS) for 2 hours in 96 well microtiter plates. EGCG (0.25mg/mL) was then mixed with dilutions of CSP (0-100 μ g/mL) in TSBS and added to the ComC- wells. Controls included wells without EGCG. The biofilm was read using a crystal violet assay after 24 hr incubation. Results: The ComC- mutant incubated with CSP was found to significantly ($p < 0.05$) exhibit more biofilm formation than wells without CSP. However, EGCG was demonstrated to have a significant inhibitory effect on the ComC- mutant. The 100 μ g/mL CSP concentration without EGCG demonstrated the highest enhancement of biofilm compared to the corresponding wells with EGCG (72% increase). In addition, the 0 μ g/mL CSP control without EGCG had 19% greater biofilm than wells with polyphenol. Conclusion: It is known that CSP is a biofilm agonist for the ComC- mutant and the EGCG is an antagonist. However, now it can be stated that EGCG in combination with CSP demonstrated a significant decrease in biofilm formation. These results suggest that a polyphenol supplement containing EGCG may be a possible way to decrease the biofilm formation of *S. mutans* in patients with severe caries risk.

P24 Effect of Nicotine on Planktonic and Biofilm Growth Phases of an Experiment. V. GUPTA*, R.L. GREGORY (Indiana University School of Dentistry)

Tobacco and cigarette smoke increase the risk of periodontal disease, one of the most widespread human diseases. It has been established that *Porphyromonas gingivalis*, a gram negative anaerobic bacterium, is one of the main causative agents of periodontal disease. Prior research indicates that *P. gingivalis* binds to *Fusobacterium nucleatum* in oral biofilms. It is not yet understood if nicotine, a major component of cigarette smoke, affects the growth of bacteria differently if added in the planktonic phase, defined as the primary subculture from agar to broth before the start of a biofilm formation experiment, or the biofilm phase, defined as the secondary subculture from broth culture to a microtiter plate. Therefore, the main objective of this study is to understand this methodological difference. *F. nucleatum* and *P. gingivalis* were both grown in anaerobic GasPak containers on blood agar plates. The media for primary subculture consisted of a Brain Heart Infusion (BBL) broth supplemented with 5 g/L yeast extract and 5% vitamin K & hemin serum at 37°C. *F. nucleatum* was subcultured in the absence of nicotine and plated on a 96 well plate to establish biofilm. *P. gingivalis* was subcultured in varying concentrations of nicotine and subcultured on top of the *F. nucleatum* biofilm. Biofilm mass was analyzed using the crystal violet technique and samples were measured in a spectrophotometer at 490 nm. The results demonstrated a statistically significant increase in biofilm formation when *P. gingivalis* was subjected to a higher nicotine concentration in the planktonic phase in comparison to a lower nicotine concentration in the biofilm phase. This data suggests a nicotine assisted activation of receptors on the surface of *P. gingivalis* specific for binding to *F. nucleatum*. Further testing on the receptors through a biotinylation assay will confirm the results.

P25 Effects of Fulvic Acid on Biofilm Formation and Human Cells. E. PARKER^{1*}, R.L. GREGORY², L.J. WINDSOR², B. ALAVANJA² (¹Indiana University-Purdue University Indianapolis, ²Indiana University School of Dentistry)

Shilajit is a traditional medicine used to treat numerous health conditions, including bone/cartilage repair and regeneration. Prior research suggests that a major active component of shilajit- fulvic acid- may reduce bacteria in the oral cavity. Because fulvic acid may inhibit bacteria and shilajit has been shown to stimulate connective tissue repair, the effect of fulvic acid on the caries-forming biofilm bacterium, *Streptococcus mutans*, and on gingival fibroblast cells, which mediate connective tissue in repair/regeneration in periodontal disease, was examined. The goal of this research was to determine whether repeated short-term applications of fulvic acid to *S. mutans* biofilm reduced the amount of established bacteria and to find the concentration of fulvic acid that may inhibit gingival fibroblast growth. In the bacterial study, *S. mutans* biofilm was grown, and 8 different dilutions of fulvic acid were applied to the same biofilm groups for 10 minutes each day over a 3-day period. Upon crystal violet staining, the optical density of the wells was obtained using a spectrophotometer. Higher fulvic acid concentrations demonstrated greater inhibition of *S. mutans* biofilm formation. 0.04% repeated applications of fulvic acid resulted in a 2-fold decrease in *S. mutans* biofilm formation, which is not observed with one application. In the study with gingival fibroblasts, cell toxicity and proliferation were examined utilizing LDH and WST-1 assays, respectively. It was determined that an 0.5% solution of fulvic acid had no effects on cell viability and proliferation. This concentration will be used to examine the effect of fulvic acid on the expression of matrix metalloproteinases (MMPs) from gingival fibroblasts, since MMPs are involved in tissue degradation and repair. This study demonstrates that fulvic acid has significant inhibiting effects on *S. mutans* biofilm formation and is safe for application to human fibroblasts within the oral cavity, up to a certain concentration.

ORAL DISEASE PREVENTION & DIAGNOSIS

- P26 Determining the Effects of Fulvic Acid on Biofilm/Planktonic *Streptococcus Mutans* Growth.** M. BOTROS^{1*}, R.L. GREGORY² (¹Purdue School of Science, Indiana University-Purdue University Indianapolis, ²Departments of Oral Biology/Pathology and Laboratory Medicine, Indiana University Schools of Dentistry and Medicine)

Fulvic acid, a major organic compound extract of Shilajit has been the focus of dental research for a while. Shilajit, a sticky tar-like substance of dark brownish color, that is used today as a traditional method in India to aid with curing bone/cartilage diseases. This experiment determined the minimum inhibitory concentration (MIC), this experiment also determined the minimum bactericidal concentration (MBC). A 3-day procedure to determine the growth vs inhibition of the *S. mutans* was conducted and bacterial readings were recorded using a spectrophotometer after treating *S. mutans* with 10% formaldehyde, crystal violet stain, and iso-propanol with 30-45 minute incubations between each. A solution comprised of a 5% concentration of fulvic acid killed all of the *S. mutans*; 5.00%, 2.50%, and 1.25% fulvic acid concentrations had bacterial absorbance of 0.000, 0.009, and 0.027, respectively, as compared to the control group's absorbance of 0.254. Additionally, solutions ranging from a two-fold dilution of fulvic acid to six-fold dilution of fulvic acid inhibited the growth of *S. mutans*. A similar trend was also observed in planktonic and biofilm formation. For all of the above, in the seventh and eighth dilution (0.078% and 0.039% respectively) of the fulvic acid, the growth of *S. mutans* bacteria was similar to the control group due to the level of dilution. Overall it was observed that fulvic acid is able to kill bacteria in strong concentrations. Additionally it is able to inhibit further growth of bacteria in lower concentrations, but once the solution becomes too dilute, it does not have an effect on bacterial growth. This data can be utilized for further research on oral bacterial growth inhibitors. Furthermore, the data collected here is a significant starting point for research on the specific minimum concentrations of fulvic necessary to inhibit oral bacteria growth in the body.

- P27 Interaction Between Tin/Fluoride-containing Solutions and Acquired Pellicle on Erosion *In-vitro*.** A. ALGARNI^{1*}, W.SIQUEIRA², F. LIPPERT¹, D. ZERO¹, A.T. HARA¹ (¹Indiana University School of Dentistry, ²The University of Western Ontario)

Although F and Sn are anti-erosive agents; their interaction with the acquired pellicle (AP), a clinically relevant factor, is not well known yet. This study aimed to compare the protective effect of Sn- and/or F-containing solutions on enamel and dentin erosion prevention and to analyze the protein profile of the Sn- and F-treated AP. Phase I tested four solutions: SnCl₂/NaF, NaF, SnCl₂ and deionized water (DIW) (negative control). Forty polished bovine enamel and dentin specimens were prepared and randomly allocated to four groups (n=10). Specimens were incubated in clarified human saliva (CHS) for 24-h for pellicle formation and were then subjected to a cycling procedure that included 5-min erosive challenges (0.3% citric acid, pH 2.6); 2-min treatment with the test solutions (between 1st, 3rd and 6th cycles); 2-h immersions in CHS, and overnight immersions in CHS. Cycles were repeated 6x/day for 5d. Surface loss (SL) was determined profilometrically. Phase II: Thirty-two polished bovine enamel specimens (n=8) were similarly prepared and incubated in saliva for 24-h and treated with the same solutions used in phase I for 2-min followed by CHS immersions for 2-h. This cycle was repeated 3x for 1d. The AP formed on the specimens were collected, and analyzed for specific protein content using liquid chromatography electrospray ionization tandem mass spectrometry (LCESI-MS/MS). For enamel, SnCl₂/NaF, SnCl₂, NaF solutions provided 89%, 67%, and 42% SL reduction respectively compared with control, while in dentin

they provided 60%, 23%, and 36%, respectively (all $p < 0.05$). Seventy-two common proteins were identified in AP of all groups, 30 exclusive to DIW, 20 to SnCl₂/NaF, 19 to NaF, and 13 to SnCl₂. SnCl₂/NaF increased the abundance of pellicle proteins more than each one alone. Combination of F and Sn ions provided the best anti-erosive effect. Composition of AP changes with different treatment solutions.

ORAL/MAXILLOFACIAL SURGERY

P28 The Use of Simulation in Pre-doctoral Medical Emergencies Training. D. VAJDA*, J. BENNETT, L.M. ROMITO (Indiana University School of Dentistry)

Objective: This purpose of this study was to determine the level and method of teaching medical emergencies management in U.S. pre-doctoral programs, particularly the extent to which patient simulation techniques are employed for instruction and /or assessment. Methods: In the fall of 2013, a 22-item survey instrument was developed, piloted with a small cohort of oral and maxillofacial surgery faculty, and modified based on feedback. Survey items assessed curriculum content and teaching methods used for medical emergencies training, and program demographics. A link to the survey was sent to the institutional email of the Academic Dean and Oral and Maxillofacial Surgery Department Chair at each U.S. dental school. The internet-based survey platform Qualtrics was used to record responses and track non-responders. Two follow-up reminders were sent via email to non-responders. Response data were de-identified and statistical analyses were conducted. Results: Completed surveys were returned by respondents from 40 schools (62.5% response rate). Of responding schools, 95% (38) offered medical emergencies training; teaching methods included lecture, seminar, and small group learning. Median instruction time was 12 hours. Of responding schools, 12 reported providing management of medical emergencies instruction via high fidelity patient simulation (HFPS), 16 used role playing, 5 employed computer-based programs, and 6 utilized standardized patients. While 6 schools reported employing HFPS for 3-5 yrs., no school reported using it for > 5 yrs.; however, 4 schools reported utilizing role-playing for > 5 yrs. While class size was not significantly associated with use of HFPS, cost was significantly associated with non-use of HFPS ($p=0.0274$). Conclusions: Although the vast majority of pre-doctoral dental programs educate students in the management of medical emergencies, few programs utilize simulation as an instructional method.

ORTHODONTICS/IMAGING/CRANIOFACIAL

P29 Cervical Vertebrae Angulation: Effects on Airway Dimensions in Orthodontic Patients. S.J. RYAN^{1*}, K.S. KULA¹, G. ECKERT², A. GHONEIMA¹ (¹Indiana University School of Dentistry, ²Indiana University School of Medicine)

Objective: The purpose of this retrospective radiographic study was to determine the relationship between craniofacial morphology, cervical vertebrae angulation, and airway dimensions in orthodontic patients. Methods: Pre-treatment cephalometric measurements of airway dimensions and cervical vertebrae angulations were recorded for 49 Caucasian female patients (mean age 17.81±8.57 years). P-value and Pearson correlation coefficients were calculated to assess the effect of cervical vertebrae angulations on airway dimensions in subjects with different craniofacial morphology. Intraclass correlations (ICC) were performed on duplicate measurements of 10 lateral cephalograms after a two-week interval to assess reliability. Results: ICC values were >0.82 for all measurements. Pearson correlation coefficients were categorized based on confidence level as follows: high (>0.75 or <-0.75), moderate (0.5 to 0.74 or -0.5 to -0.74), or weak (0.3 to 0.49 or -0.3 to -0.49). A high correlation (0.83) was observed between the dimension of the bony nasopharynx (Ba-PNS) and the position of the atlas vertebrae (AA-PNS).

Moderate correlations were observed between PNS-ppw1 (0.59) and PNS-ad1 (0.58) with regards to the position of the atlas vertebrae (AA-PNS). A moderate correlation was also observed between the oropharyngeal airway dimension (PNS-ppw2) and the following cervical vertebrae angulation parameters: CVT-SN (0.51), OPT-SN (0.55), and OPT-RL (0.52). Conclusions: As the atlas vertebra (CV1) was positioned further from the posterior maxilla, the size of the nasopharyngeal airway appeared to increase. As the angle between the cranial base and CV2 and CV4 increased, the size of the oropharyngeal airspace appeared to increase. The A-P position of CV1 and the angulations of the cervical vertebrae at CV2 and CV4 relative to the cranial base affect the upper airway dimensions. The amount of change of the upper airway size was variable with the extent of the cervical vertebrae angulations. (This project was supported by the IUPUI 3D Craniofacial Complex Imaging Center; Jarabak Professorship)

P30 Vitamin A Metabolic Defect Impairs Maxillary Arch and Incisor Development. P.

SANGOI*, D. HADEL, T. SALTER, R. LINDMAN, L. SANDELL (University of Louisville School of Dentistry, Department of Molecular, Cellular, and Craniofacial Birth Defects)

Objective: To determine if mice with defect in Vitamin A metabolism have abnormal tooth development or microstomia (small mouth), characteristic of many human craniofacial syndromes. Methods: Mouse embryos with a mutation of the Rdh10 gene have a deficiency of Vitamin A metabolism that is lethal prior to tooth formation. The diet of pregnant female mice were supplemented with partially metabolized Vitamin A to ensure survival of mutant embryos until embryonic day 14.5 (E14.5). Frontal and transverse embryo sections were prepared and stained with Hematoxylin & Eosin. Cryosectioning was conducted and cell apoptosis was assessed. Sections were analyzed using both light and fluorescence microscopes. Quantitative measurements were taken using Image J software. Results: Incisor morphology is abnormal in Rdh10 mutant embryos. The primary enamel knot, a signaling center that regulates tooth cusp morphogenesis, is absent in mutant embryo incisors. Abnormal incisor morphology was not due to differences in cell apoptosis. Mutant incisors have a smaller aggregation of neural crest derived mesenchymal cells that form the dental papilla. The most significant difference was the size of the maxillary arch. Measurements indicate that maxillary arch area in mutant embryos was 79% that of wild type. Conclusion: Mice with a disturbance in Vitamin A metabolism have abnormalities in development of incisors. Mutant embryos also display microstomia, with the maxillary arch being significantly smaller than wild type embryos. Results suggest that Vitamin A metabolism deficiency in humans could be responsible for cases of craniofacial defects such as microstomia and abnormal tooth morphology.

P31 Epigallocatechin Gallate Treatment Affects Ts65Dn Craniofacial Airways and Morphology.

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Down syndrome (DS) is caused by having three copies of chromosome 21 (i.e. Trisomy 21). DS is associated with many detrimental phenotypes including intellectual disabilities, heart defects, and abnormal craniofacial development. Additional complications common in individuals with DS arise from this altered craniofacial development such as obstructive sleep apnea, which is associated with restricted oronasal airways and an underdeveloped mandible. Ts65Dn mice are trisomic for about half of the orthologs on human chromosome 21 and display many phenotypes associated with DS including craniofacial abnormalities. Dyrk1a is found in three copies in Ts65Dn mice and individuals with DS. Dyrk1a is thought to play a key role in craniofacial morphogenesis. Epigallocatechin gallate (EGCG) is a green tea polyphenol and inhibitor of Dyrk1a activity. We hypothesize that decreased Dyrk1a activity in Ts65Dn mice will ameliorate craniofacial dysmorphology. To test our hypothesis we compared Ts65Dn mice, Ts65Dn mice treated with EGCG, and Ts65Dn mice without an extra copy of Dyrk1a. Six week old

mice were sacrificed and their heads imaged using micro-computed tomography. From the images, we measured nasopharyngeal airway volumes and anatomical landmarks (n = 54) to assess local differences in craniofacial and mandibular morphology between samples. Our preliminary results indicate that EGCG treatment and reduced Dyrk1a copy number increases mean nasopharyngeal airway volume in Ts65Dn mice. Craniofacial morphometric differences were found among all samples. Patterns of variation suggest that both EGCG treatment and reduced Dyrk1a copy number affects craniofacial morphology.

P32 Web-Based Interactive Instruction in Intraoral and Panoramic Radiographic Anatomy. S. ALMASSI*, A. FARKHONDEH, J.R. GEIST (University of Detroit Mercy School of Dentistry)

Objective: Evaluate the effectiveness of web-based interactive modules in the instruction of dental hygiene students on intraoral and panoramic radiographic landmarks. Methods: The class of 2013 (experimental group, n=23) was assigned to study radiographic landmarks as presented on interactive web-based modules, produced by the Michigan Dental Association, instead of classroom presentations. The intraoral module (IOM) consists of radiographs on which landmarks are illustrated, with narration describing each landmark. Two interactive quizzes are included for student self-evaluation. The panoramic module (PNM) uses a panoramic radiograph marked with points representing landmarks. Students place the cursor over each point to learn the name of the landmark. Knowledge of landmarks was assessed through one in-class "open-book" project and five "closed-book" quizzes and examinations. Unpaired t-tests compared the experimental group with the previous year's class (control group, n=24), which had received instruction in the traditional classroom format. A survey was administered to the experimental group to determine their perceptions of instruction with the modules. (IRB Approval #1314-56). Results: The experimental group scored significantly higher than the control group on the project ($p < 0.001$). On quizzes and examinations, experimental group scored lower than control group for both types of landmarks, while differences for intraoral landmarks were not significant ($p = 0.072$), the differences for panoramic landmarks were significant ($p = 0.039$). 30% of the students preferred computer-assisted instruction to classroom instruction. Conclusions: Narration and interactive quizzes on the IOM may have contributed to the similar performance of experimental and control groups, while their absence may have adversely affected the effectiveness of the PNM. The modules may serve as a reference source for study, but may not be as effective or as well received as a primary instructional tool.

P33 Airway Dimensions and Pathologies of Trumpet Players vs. Non-trumpet Players. K.S. KULA^{1*}, A. IMBURGIA¹, S. HALUM², M. VAN DIS¹, A. GHONEIMA¹ (¹Indiana University School of Dentistry and School of Medicine, ²Indiana University Purdue University at Indianapolis)

Objective: The objective of this retrospective, 3-dimensional cone beam computed tomography study was to determine if there is a significant difference between the most constricted area of the airway, the prevertebral soft tissue thickness and airway dimensions (length and volume) of the nasal cavity, nasopharynx, oropharynx, and maxillary sinuses of university trumpet players versus non-trumpet playing controls. The second objective was to determine significant differences in the prevalence of airway pathologies between university trumpet players and controls. Method: Following IRB approval (#1102-23) and consent and reliability studies, measurements of airway parameters and pathology were compared between 66 Caucasian trumpeters and 22 ethnic-matched controls. An analysis of covariance, with age and sex included as covariates, was used to compare the airway measures. Since there was a significant difference in gender and age, comparisons between groups for the presence of any airway pathologies was made using logistic regression including age and sex as covariates. A 5% significance level was used for all comparisons. Result: The trumpet players had significantly smaller nasal cavity volume ($18028 + 595 \text{ mm}^3$ vs. $25266 + 1116 \text{ mm}^3$; $p < 0.0001$) and significantly greater soft tissue thickness at

CV2ia (3.29 + 0.10 mm vs 2.70 + 0.10 mm; p=0.03) and CV3sa (4.55 + 0.13 mm vs 3.74 + 0.14 mm; p=0.005) than the controls. No other airway measure demonstrated a significant difference between the two groups. Trumpeters had significantly (p=0.002) more airway pathology (n=33; 50%) than the controls (n=4; 18%). Antral pseudocysts or polyps composed 52% of trumpeter pathologies as compared with 0% controls. Conclusion: The only significant differences in airway dimensions between trumpet players and controls were decreased nasal cavity volume and some areas of prevertebral soft tissue thickness. Trumpeters were almost three times as likely to exhibit airway pathology when compared with controls. (Funding: IUPUI Signature Center Initiative – 3D Imaging of the Craniofacial Complex Center and the Joseph and Aida Jarabak Endowed Professorship)

P34 Facial Soft-Tissue Depth Asymmetry and Surgically Corrected Unilateral Clefts. J. STARBUCK*, A. GHONEIMA, K.S. KULA (Department of Orthodontics and Oral Facial Genetics, Indiana University School of Dentistry)

Cleft lip with or without cleft palate (CL/P) is a relatively common craniofacial malformation that disrupts bony and soft-tissue development of the labionasal facial regions. Individuals with CL/P typically undergo multiple surgeries to close the cleft and improve appearance of the cutaneous upper lip and nose. The combination of CL/P and subsequent craniofacial surgeries can cause scarring and muscle pull and can result in soft-tissue depth asymmetries across the face. We tested the hypothesis that bilateral facial tissue depths of children with surgically repaired unilateral CL/P exhibit differences in symmetry. Following IRB approval (study # 1210009813), reliability studies were carried out to assess intra-class correlation (ICC) and technical error of measurement (TEM). A total of 28 bilateral tissue depths were measured by one investigator on cone beam computed tomography (CBCT) images from orthodontic records of children with unilateral CL/P (n = 55) who have been surgically repaired, aged 7-17 yrs., using Dolphin software (v 11.5). Paired t-tests were used to determine whether the tissue depths on each side of the face were significantly different, and a p-value of ≤ 0.05 was considered significant. ICC was high (0.99), indicating that tissue depth measurements were reliable. TEM was low (<0.20 mm), indicating that measurement error is acceptable. Significant differences in tissue depth symmetry were found around the cutaneous upper lip and nose, with tissue depths on the clefted side being significantly increased by (0.6 - 1.6mm). Despite the best efforts of plastic surgeons there is residual facial asymmetry from CL/P, although these asymmetries are small. Surgeons can quantify asymmetry using 3D CBCT images and 3D imaging software to develop individualized treatment plans for patients after evaluating the anatomical relationship between the hard- and soft-tissues of the craniofacial complex. (Supported by Indiana University Signature Center Initiative: 3D Imaging of the Craniofacial Complex Center; Jarabak Endowed Professorship)

P35 A Novel Approach to Assessing Orthodontic Bracket Placement Teaching Techniques. T. LUEDERS*, A. GHONEIMA, K. STEWART (Indiana University School of Dentistry)

The skill of orthodontic bracket placement is a central concept of any orthodontic residency clinical curriculum, yet there are no documented studies that investigate the most effective method of teaching this indispensable clinical skill. Purpose: The purpose of this pilot study was to assess the impact of different pedagogical modalities on the development of bracket placement abilities in orthodontic graduate students using a novel digital model superimposition technique for evaluation. Methods: Orthodontic graduate students with less than one year of bracket placement experience practiced bracket placement on either hand-held models or models mounted in a simulation mannequin to determine which method of instruction was more effective in producing accurate bracket placement. Thirteen models were scanned using the iTero 3D scanner and bracket placement was evaluated using 3dMD Vultus software to superimpose the models digitally and quantify their difference from the gold standard. All data were

analyzed using mixed ANOVA and reliability tests were done using intra-class correlation coefficient (ICC). $P < 0.05$ was considered statistically significant. Results: ICC values were > 0.89 for all measurements. Results from the distance measurements of each individual bracket to specified landmarks on the cast were not statistically significant. However, results from the superimposition assessment, where two casts were digitally superimposed and their difference quantified, were statistically significant. Although both groups demonstrated statistically significant improvements in bracket placement accuracy following the training, the group that trained on the mounted models had a greater improvement in accuracy ($p < 0.05$). Conclusion: This pilot study defines an optimal method of bracket placement instruction. Additionally, a digital model superimposition technique is introduced as an accurate method of providing objective feedback to orthodontic graduate students throughout their clinical training. (Supported by IUPUI Program Review and Assessment Committee (PRAC) grant)

P36 Maxillary Mucosal Thickening in Unilateral Cleft Lip and Palate. L. HALE^{1,2*}, J. STARBUCK¹, A. GHONEIMA¹, K.S. KULA¹ (¹Department of Orthodontics and Oral Facial Genetics, Indiana University School of Dentistry, ²Department of Mathematical Sciences, Indiana University-Purdue University Indianapolis)

Cleft lip and palate (CLP) perturbs nasolabial development, often resulting in chronic maxillary sinusitis associated with maxillary sinus mucosal thickening (MT). This study quantifies maxillary sinus MT in children born with unilateral CLP. We hypothesize that MT is increased in children with CLP relative to controls. We define "MT" as the difference between the entire maxillary sinus volume and airspace volume. Cone beam computed tomography (CBCT) images of 8-14 yr. old age- and sex-matched unilateral CLP patients ($n = 10$) and controls ($n = 10$) were obtained (IRB approval # 1210009813). Maxillary sinus and airspace surface areas (SAs) were measured on individual CBCT slices in coronal view. SA measurements were summed and multiplied by voxel size (0.4mm) to obtain a volume. Paired t-tests determined whether maxillary sinus volume, air volume, MT (i.e. maxillary sinus volume – airspace volume), and percentage of MT (i.e. MT/maxillary size $\times 100$) differed. A p-value of ≤ 0.05 was considered significant. Intra-class correlation assessed reliability and was high (0.99). Significant differences were found for several measurements: Maxillary airspace (non-cleft side vs. right side control $p\text{-value} = 0.002$; cleft-side vs. left side control $p\text{-value} = 0.004$), MT (cleft-side vs. left side $p\text{-value} = 0.009$), and percentage of MT (non-cleft side vs. right side control $p\text{-value} = 0.002$, cleft-side vs. left side control $p\text{-value} = 0.002$). Maxillary airspace was decreased by 30% (non-cleft side) and by 33% (cleft side). Percentage of average MT was 40% (non-cleft side) and 42% (cleft side) of CLP patients, but only 9% (left and right side) in controls. Surgically repaired CLP patients exhibit decreased maxillary airspace and increased MT relative to controls. CLP deformities are associated with MT, and 3D imaging is useful for quantitatively evaluating MT. (Funding: IUPUI Signature Center Initiative – Three Dimensional Imaging of the Craniofacial Complex Center; Jarabak Endowed Professorship)

PEDIATRIC DENTISTRY

P37 Referring Child Patients: A Pilot Survey of Indiana Pediatric Dentists. Z. BOZIC*, J.E. KOWOLIK (Indiana University School of Dentistry)

Dental caries is one of the most common chronic diseases affecting children in the United States with a majority of children having one or more carious lesions before age five. General dentists may be a child patient's first exposure to dental care and they make referrals to pediatric specialists for a variety of reasons. The object of this study was to evaluate the frequency of, and reasons for, referral of child patients from general dentists to pediatric dental specialists. Using an IRB approved (#1307011757) phone survey of the active members of the Indiana Society of Pediatric Dentists, participants were

informed of the goals of this study and read the questionnaire using a pre-determined script. No more than three attempts were made to contact each participant and dentist responses were de-identified and organized using a numerical coding system. Participating specialists indicated receiving 65% of referrals based on the child patient's behavior. Results suggest that a majority of pediatric dentists receive over five referrals per month and a majority of their referrals are based on the behavior of the child patient.

P38 Chart Review of Treatment of Retained Primary Mandibular Molar Teeth. A. SCULLY*, J.E. KOWOLIK (Indiana University School of Dentistry)

There exists an insufficiency of reported research related to retained primary teeth in the adult mouth. Studies that exist lay a foundation but have not extended past 25 years. Objectives: This study aims to calculate the age that the primary teeth remain in the mouth or were lost, if males or females have a longer expected lifespan of the teeth and if there is a difference between the expected lifespan of the teeth based on race or payment method. Investigating these questions will give us a better understanding of how long we can expect these retained teeth to remain in the mouth, if that expectation varies based on certain demographic information and if future studies should investigate older subjects. Methods: This study was approved by the IU-IRB (1306011688). AxiUm® electronic medical software was used to access previously recorded patient records. The inclusion parameter is presence or previous presence of #K or #T. The exclusion parameter is a subject <15 years. 75 charts are selected and de-identified. Data points include age, gender, race, payment method and craniofacial history. Survival analysis is used to estimate the median age of retained #K or #T and the median age at which these teeth were lost. Cox proportional hazards are used to test the effects of gender, race and payment method. Results/Conclusions: 97 teeth from 75 subjects were analyzed. The subject's age at the time of the study was 34±11 years with a range of 15-67 years. 29% of the teeth were not retained in the mouth and demographic data had no statistical significance on outcome. The estimated mean survival of the teeth is 47 years. A large age range of patients exist with retained #K or #T. 47 years far surpasses the age in current literature and future studies should include older subjects.

P39 What Do Adults Know About Baby Teeth - A Pilot Study. L. DESANTIS*¹, J.E. KOWOLIK¹, G. ECKERT² (¹Indiana University School of Dentistry, ²Indiana University School of Medicine)

The purpose of this study was to identify the general knowledge and the understanding of the adult population regarding the sequence and timing of the eruption and exfoliation of the primary dentition. The study was completed at Indiana University School of Dentistry. Four hundred randomly identified self-selected adults attending the Comprehensive Care Clinic or accompanying their children for dental care at the Pre-doctoral Pediatric Clinic and the Graduate Orthodontic Clinic were surveyed. A comprehensive, closed-ended, self-administered questionnaire was employed. Responses were collated and entered into a computer database and examined statistically using chi-square tests. Overall, 96% of the population believed that primary teeth are important, however only 26% believed that the caregiver should brush a child's teeth 3 times a day. 63% of the surveyed population did not correctly answer the location for the eruption of the first primary tooth, and 42% believed the location varied from child to child. 38% believed that all primary teeth have erupted by 2 years, while another 38% believed that it was 5 years. 55% of the population believed that the primary canine erupts before the primary first molar. 80% believed that the primary canine exfoliates before the exfoliation of the first primary molar. 32% believed the first primary molar exfoliates between the ages of 7-9, whereas 32% of the population believes that the first molar exfoliates between the ages of 10-12. 37% believed that any one of their child's primary teeth could be lost naturally at any age, and 15% believed that there was no pattern at all for tooth eruption. Adults with only some level of high school education were more likely to strongly agree that teeth can exfoliate naturally at any age for a child (p=0.0416). The Hispanic and Asian population were also more likely to

strongly agree that any tooth could exfoliate naturally at any age ($p=0.0113$). The 70+ population answered that caregivers should only brush the child's teeth 1x per day ($p=0.0025$), and were also very unlikely to agree that there was a pattern to the exfoliation of primary teeth ($p<0.0001$). These results reveal a deficit in the overall knowledge on primary dentition in this sample. To improve oral health it is necessary to promote oral health awareness and understanding on the eruption and exfoliation of primary dentition. This study presents an insight into an otherwise unknown domain and illustrates the need for focused education.

PERIODONTICS

P40 Response of Salivary Epithelial Cells to Periodontopathic Bacteria. C. BLACKBURN*, R.L. GREGORY, and M. SRINIVASAN (Department of Oral Biology and Oral Pathology, Medicine and Radiology, Indiana University School of Dentistry, Indianapolis, IN)

Chronic periodontitis is the most prevalent form of inflammatory periodontal conditions initiated by sequential colonization of a broad array of bacteria and perpetuated by host response to the changing biofilm. Disease-associated bacteria, including *Porphyromonas gingivalis*, *Fusobacterium nucleatum* and *Aggregatibacter actinomycetemcomitans*, have been detected at high frequency in oral mucosa covering the cheek, tongue and palate. Significantly, treatment does not affect the prevalence of periodontopathic bacteria on oral mucosal epithelial cells implicating the oral mucosal epithelial cells as potential reservoirs that facilitate re-infection. Previously, over 40% of epithelial cells in unstimulated whole saliva have been shown to be viable and capable of responding to microbial ligands. In this study we investigated that the response of the epithelial cells in saliva to periodontal pathogens could play a role in the persistence and/or recurrence of chronic periodontitis. Salivary epithelial cells (SEC) isolated from healthy saliva and those from patients with chronic periodontitis were stimulated with the periodontal pathogens *P. gingivalis* and/or *F. nucleatum* as biofilm or planktonic formulations in varying proportions for 4 to 24hrs. Culture supernatants were assessed for cytokines by ELISA. In general the SEC secreted higher amounts of IL-8 in response to *P. gingivalis* than that to *F. nucleatum* alone or a combination of *F. nucleatum* and *P. gingivalis*. A wide variation was observed in the response of the SEC from different healthy saliva samples to the periodontal pathogens. Conclusion: The response of the SEC to periodontal pathogens could lead to better understanding of the nature of recurrence of chronic periodontitis.

P41 Peptidoglycan Recognition Proteins: Expression Profile in Chronic Periodontitis. S. MAKKATTIL*, P. SHARMA, V. JOHN, S. PRAKASAM (Indiana University School of Dentistry)

Objective: Pattern recognition receptors (PRRs) play important roles in periodontal disease progression. Peptidoglycan recognition protein (PGRP), a novel class of PRR, are expressed as downstream of TLRs and NOD. Four types of human PGRP have been reported with distinct functions and cellular expression. PGRP1, PGRP3, and PGRP4 are bactericidal and are anti-inflammatory. PGRP2 is a pro-inflammatory alarmin. Previously, we showed that PGRP3&4 are reduced in shed epithelial cells in saliva of subjects with chronic periodontitis (CP). Uehara et al demonstrated that oral epithelial cells respond to ligands of TLRs and NODs by up regulating PGRP without concomitant release of inflammatory cytokines. The objective of this study was to examine the expression profile of PGRP in diseased gingival tissues of CP patients and compare it with tissues from subjects with healthy gingiva (H). Methods: A cross sectional study design was used. In the test group (CP), diseased tissues were collected from 10 subjects with generalized severe CP and in control group (H) healthy tissues were collected, from 10 subjects. Tissues were analyzed with qPCR (PffafI method) and immunohistochemistry to estimate and compare expression of PGRPs, TLR2, TLR4, NOD1, and NOD2. Mann Whitney U test was used to test statistical

significance. Correlation Coefficients between the different PRRs were also estimated. Results: In gingival tissue of subjects with CP, levels of PGRP2, PGRP3, and PGRP4 were increased significantly ($p \leq 0.01$) when compared to H. PGRP1 levels were also increased but statistically equivocal. Interesting correlations were noted between the expression patterns of different PRRs. Conclusions: This is the first report documenting increased expression of PGRPs in tissues from sites with CP. Understanding the role of PGRP in CP may shed additional light on its complex pathogenesis. Moreover, PGRP may serve as biomarkers for periodontal disease.

PRACTICE MANAGEMENT/ PUBLIC HEALTH

P42 Practice Management Confidence and Satisfaction Levels in Practicing Dentists. A. MOUKLED*, F. SHOUNIA, M. WHEATER, M. AKSU (University of Detroit Mercy School of Dentistry)

Objective: The purpose of this survey was to determine attitudes of practicing dentists toward dental practice management and their recollection of the adequacy and level of satisfaction of their practice management experience during dental school. Methods: A 16 question survey was administered to dentists during the 2013 annual meeting of the Michigan Dental Association. This study was approved by the UDM Institutional Review Board (Protocol #1213-80). Results: Sixty-five responses were collected. Forty-one respondents graduated more than 20 years ago and 40% reported disliking the business aspect of the practice. 15% reported they were insecure in their ability to run the business of the practice. Of the respondents with 15 or fewer years since graduation, 14.3% disliked the business aspect of practice, and 4.8% were insecure in their ability to run the business of the practice. Of those dentists with 15 years or less since graduation 47% agree with the statement that “they did not appreciate the relevance of practice management while in dental school,” compared to 41% of respondents who graduated more than 20 years ago. 52% of the respondents with 15 or fewer years since graduation report that the amount of time was inadequate. Of those with more than 16 years since graduation, 72% thought that the time devoted to practice management was inadequate. Of those with 15 years or less since graduation, 75% enjoy spending time taking practice management courses as compared to 39% of those who graduated more than 20 years ago. Conclusion: It appears that the complexity of business ownership increases as time since graduation passes, and the perception of the adequacy of the curriculum decreases over time.

P43 Healthy Smiles for Employability – An Oral Health Program. J. BHAHEETHARAN*, E. SCHMIT, S. HENDRICKS, K.M. YODER, T. CARLSON (Indiana University Fairbanks School of Public Health, Indiana University School of Dentistry, Office for Civic Engagement and Health Policy)

The Indiana University Student Outreach Clinic is a free student-run clinic that provides medical, pharmacy, legal, dental, social work, and physical therapy services at a community church in Near Eastside Indianapolis, Indiana. The Near Eastside (NES) Indianapolis community is subject to several negative social determinants of oral health, including low income, which can impede access to oral health care and put residents at a higher risk for oral health problems. Through the IU School of Dentistry SOC, a need was identified to increase access to affordable dentures and improve employment opportunities. The Healthy Smiles for Employability (HSE) program was created to improve oral health, well-being, and employment outcomes in the NES by providing free dentures to low-income, uninsured residents, and connecting them with local agencies that provide job assistance services. Program components of HSE include (1) program organization, (2) community engagement, (3) participant recruitment and enrollment, (4) dental and employment services, and (5) program evaluation. Eligible HSE candidates include

individuals who are unemployed or underemployed (e.g. income below 200% federal poverty line), seeking to improve their job situation, and perceive the appearance of their teeth (e.g. missing front teeth) as a barrier for greater employment opportunities. Enrolled HSE participants receive dental services at the IUSD-SOC located in the NES at HealthNet People's Health and Dental Center and at the IUSD Clinic. Collaboration with community and health centers, local health providers, free clinics, homeless shelters, religious organizations, and others serves as an immense asset to recruit HSE candidates and provide job advancement and retention services, social services and other essential resources to HSE participants. Ongoing program evaluation serves to increase program effectiveness and organization in order to support the success of HSE including its community partners and participants. (Supported by the IUPUI Solution Center)

PROSTHODONTICS

P44 Replica Implant Proof-of-Concept Study. P.S. WULFF*, A. FARAG, A.G. FARMAN, W.C. SCARF, T.J. GORNET, G.E. GRAF, L. GETTLEMAN (University of Louisville)

Objectives: Determine the feasibility of making a replica implant of a periodontal ligament-attached tooth using current technology, in fresh hog mandibles and later humans. Clinical indications: a) vertical cracks and horizontal tooth fractures, b) deep caries; c) coronal caries under reusable fixed prostheses; d) root or chamber perforations; e) tooth resorption; f) endodontic failures; and/or g) lower cost alternative to endodontic treatment of the tooth, post reinforcement and crown. Methods: Cone beam computerized tomographic files of individual teeth were generated from a hog mandible using an iCAT 17-19 machine. Using 3DSlicer, a digital file of the lower right second deciduous premolar was generated, blocking out the pulp chamber and manually selecting the contrast level at the exterior of the tooth. The DICOM file was converted to an STL file for replication in an additive manufacturing process using High Temperature Selective Laser Sintering to generate Nylon 12 replicas of the entire tooth. Futures replicas may be made in metals (CoCrMo, Ti, Ti6Al4V) or ceramics. The tooth was then extracted from the hog mandible and the replica tooth immediately implanted, similar to an avulsed tooth. To confirm, pre-op and post-op x-rays were taken and replica tooth was coated in barium (for radiopacity). Results: After troubleshooting digital file compatibility and precision/accuracy of the CBCT image, the replica was successfully prototyped and implanted. Conclusion: Prototyping replica implant from CBCT was successful. After prototyping a replica implant tooth from Nylon 12, more studies will address the accuracy of this replica implant versus the tooth being replaced. The digital process should be automated. Before clinical trials, animal studies are needed to determine biocompatibility of the replica implant, verified by histological examination. Surface modification for connective tissue attachment and cervical membrane placement in animal models to prevent epithelial migration around the replica implant should be explored.

SALIVARY RESEARCH

P45 Adipokines in Saliva as Markers for Type-2 Diabetes Mellitus. A.J. RABER^{1*}, M. MOHAMMAD², M. SRINIVASAN¹ (¹Department of Oral Pathology, Medicine and Radiology, Indiana University School of Dentistry, ²Indiana University Purdue University Indianapolis)

The American Diabetes Association estimates that over 23 million adults and children in the US have diabetes; nearly 95% develop type 2 diabetes mellitus (T2DM). An additional 41 million Americans are believed to be pre-diabetic, according to the American Diabetes Association. The total economic burden of T2DM in the United States is in excess of \$132 billion annually, including \$92 billion in direct medical costs. Dysregulation of many biological pathways precedes the development of overt T2DM characterized by chronic hyperglycemia and reduced beta-cell effectiveness. Improvement of risk prediction for T2DM is

crucial for identifying high-risk individuals who could benefit from specific preventive measures. Serum proteomic and metabolite profiles suggest significant differences between patients with T2DM and healthy individuals. Elevated levels of inflammatory mediators and adipokines in serum have been suggested as predictive markers for T2DM. Saliva proteome analyses suggest that it shares greater than 20% of proteins and peptides with serum. The overall objective of this study is to determine the efficacy of salivary adipokines (resistin and visfatin) as markers for T2DM. Unstimulated whole saliva (UWS) was collected by the drooling method from individuals reporting to the clinics of the Indiana University School of Dentistry after obtaining informed consent according to the Institutional Review Board. Samples were collected from T2DM individuals who provided the HbA1c values determined within the past three months. UWS from age and sex matched healthy individuals constituted the control samples. All UWS samples were collected in pre-chilled test-tubes, place on ice immediately after collection and processed within 15 minutes. All samples were clarified by centrifuging at 4000xg at 4°C for 10 mins and stored in Complete™ Protease Inhibitor Cocktail (Roche, Mannheim, Germany) at -80°C. The levels of resistin and visfatin in clarified saliva were determined using specific assay kits (Cayman chemical company, Ann Arbor, MI). Statistical significance of the proteins between the diabetic and healthy saliva was determined by students' T- test. Although the concentration of both resistin and visfatin was higher in diabetic saliva than that in healthy saliva, the difference was significant only for the latter. In conclusion, our data suggest that the salivary visfatin could represent a potential marker for T2DM.

TISSUE REGENERATION AND REPAIR

P46 Poly-L-lactic acid/Dicalcium Phosphate Dihydrate Composite Scaffolds for Bone Tissue. N. TANATAWEETHUM*, W.C. LIU, T.G. CHU (Indiana University School of Dentistry)

Introduction: Dicalcium phosphate dihydrate cement (DCPD) has proven to be an efficient bone substitute because of their properties, such as biocompatibility and low setting temperature, which allow the incorporation of various drugs. However, DCPD cement has some drawbacks related to its low mechanical strength. To address this limitation, addition of sodium citrate as a regulator and polylactic acid (PLLA) as a reinforcing agent have been proposed in this study. Objectives: 1) To develop composite PLLA/ DCPD scaffolds with enhanced compressive strength. 2) To examine the cytocompatibility of PLLA/DCPD composite. 3) To investigate in vitro kinetic release of romiplostim (Nplate)-loaded PLLA/DCPD composite. Materials and Methods: DCPD cement was synthesized with a 1:1 molar ratio of monocalcium phosphate monohydrate and β -tricalcium phosphate. Sodium citrate and deionized water was used as liquid phase. The cement paste was prepared at powder to liquid mass ratio (P/L) of 1.50. The scaffolds were fabricated by indirect casting method. Then, they were coated with 5 % PLLA solution. The compressive strength of the scaffolds was measured. In addition, cell adhesion, proliferation and differentiation were examined by scanning electron microscopy, XTT and ALP assay kit. Nplate-loaded PLLA/DCPD composite disks were prepared to study in vitro kinetic release. The release profile was investigated at 1 h, 4 h, 8 h, 24 h, 72 h, and 168 h by ELISA kit. Results: sodium citrate and PLLA coating significantly increased compressive strength of DCPD scaffolds from 0.37 ± 0.06 MPa to 1.70 ± 0.17 MPa ($p < 0.05$). The cytocompatibility test showed that DCPD and PLLA/DCPD composite were non-cytotoxic. Furthermore, they stimulated the cell proliferation and expressed ALP activity. Conclusion: a combination of indirect casting and polymer coating methods can be applied to fabricate a cement/polymer biodegradable scaffold for bone tissue regeneration without cytotoxicity.

P47 Effect of Kalirin Domains on Alkaline Phosphatase Activity in Osteoblasts. J. KABIR*, A. MAHMOUDI, M. VANG, K. WAYAKANON, P. ELENISTE, A. BRUZZANITI (Indiana University School of Dentistry)

Osteoblasts (OBs) are important for maintaining bone formation. Disruption between the balance of OBs and osteoclasts, which are responsible for bone resorption, can lead to osteoporosis, a disease that causes a decrease in bone density and an increase in bone fragility. Disruption of the function of OBs can also lead to low bone mass. Our lab has previously published that Kalirin, a GTP-exchange factor protein, is important for regulating bone mass. Global deletion of Kalirin in mice results in osteoporosis. Kalirin plays a role in the signaling pathways of OB function, but its mechanism of action is unknown. There are three different isoforms of Kalirin: Kal7, Kal9, and Kal12. Each of the isoforms contain different functioning domains that may have an effect on the function of OBs. To further study the relationship between Kalirin and OB function, the effects of different Kalirin protein domains on the activity of alkaline phosphatase (ALP), an enzyme responsible for osteogenic activity, were examined in an osteoblast cell line, MC3T3-E1. Constructs of Kalirin were made which contained four protein domains: GEF1, GEF2, IGFN and Ser/Thr kinase. MC3T3-E1 cells were transfected with these Kalirin constructs and ALP assays were then performed on the cells. The experiments revealed that GEF2 leads to an increase in ALP activity, while the Kinase, GEF1 and IGFN domains have no distinguishable effects on ALP activity. In conclusion, since Kalirin is known to activate RhoGTPase via its GEF domains, the GEF2 domain may be increasing ALP activity by increasing Rho activity in OBs. Identifying ways to activate GEF2 may be a novel target to increase bone regeneration, which can then be applied for the treatment of bone loss associated with periodontal disease and osteoporosis.

P48 Expression and Function of Tyrosine Kinase Pyk2 on Osteoblast Differentiation. S. POSRITONG^{1*}, P. ELENISTE¹, K. WAYAKANON², A. BRUZZANITI¹ (¹Indiana University School of Dentistry, ²Faculty of Dentistry, Naresuan University, Phitsanulok, Thailand)

The balance of osteoblastogenesis and osteoclastogenesis are important for bone remodeling. Excessive osteoclastic bone resorption or decreased osteoblast (OB) function can lead to low bone mass, which may result in osteoporosis. Proline-rich tyrosine kinase 2 (Pyk2) has been shown to be an important factor in bone mass regulation. Our studies and others demonstrated the absence of Pyk2 in mice (Pyk2-KO) led to increased bone formation, resulting in enhanced bone mass. There are 2 isoforms of Pyk2, which are full-length Pyk2 and the smaller spliced-form, Pyk2-S. However, the mechanism leading to high bone mass and Pyk2/Pyk2-S expression is unclear. The objective of this in vitro study was to determine expression and function of the Pyk2 isoforms on OB differentiation. Primary OBs from wild-type mice were cultured for 0-21 days and cells were collected on day 0, 3, 7, 10, 14 and 21. OBs were also treated with ascorbic acid (AA) and β -glycerol phosphate (β -GP) to induce differentiation. After this, RNA extraction and reverse-transcription (RT)-PCR were performed to analyze Pyk2/Pyk2-S mRNA expression. In the absence of AA+ β -GP, Pyk2 expression was higher than Pyk2-S at 0, 3 and 7 day, but by 14 days, Pyk2-S mRNA expression was more than that of Pyk2. However, in the presence of AA+ β -GP, Pyk2/Pyk2-S mRNA expression disappeared within 6 hours of treatment. In other studies, Pyk2/Pyk2-S were transiently expressed in MC3T3-E1 cells and alkaline phosphatase (ALP) activity, a marker of OB osteogenic activity, was quantified. Data were analyzed using one-way ANOVA with Tukey's test ($\alpha = 0.05$). MC3T3-E1 expressing Pyk2-S showed significantly reduced levels of ALP activity compared to Pyk2 ($p < 0.001$). These results revealed differential expression of Pyk2 and Pyk2-S during OB differentiation and that Pyk2-S inhibits ALP activity. Targeting Pyk2-S may be a novel strategy to increase OBs activity and bone mass.

- P49 Exploring Biological Property of a New Dental Pulp Cell.** C. LOVE¹, Z. HUANG², Y. EHRLICH², L.J. WINDSOR², F. SONG² (¹Department of Biology, School of Science, IUPUI, ²Indiana University School of Dentistry)

Stem cells have introduced a brand new strategy to the field of regenerative medicine. Although many types of stem cells have been found, the limitations of each type of stem cells demand the exploration of new cell line. Here we report a new dental stem cell line recently developed from a natal tooth on its adipogenesis property. Upon reaching 100% confluence, neonatal pulp cells were cultured with 4.5 mg/ml glucose conditions followed the published protocol. A commercial human pulp stem cell line was used as a comparison. Both cell lines were stained with Oil Red O stain after 16 and 30 days of induction. The observations of fat content were recorded and compared. The preliminary data indicated that natal tooth pulp cells have the ability to differentiate into adipocyte, when the right environmental factors are provided.

TOBACCO RESEARCH

- P50 Assessing the Depiction of Electronic Cigarettes in YouTube Videos.** R. HURWICH*, L.M. ROMITO (Indiana University School of Dentistry)

Objective: Use of electronic cigarettes is an emerging phenomenon with unknown health implications. YouTube videos of e-cigarettes are abundant, easily accessible, and possess the potential to promote e-cigarette use through the increased visibility of user-generated content and novel marketing strategies. As there are no published studies examining the content of e-cigarette videos, this study aimed to assess the depiction of e-cigarettes in terms of content and viewing qualities. Methods: Two calibrated reviewers conducted 4 searches for e-cigarette English language YouTube videos between August 2, 2013 and November 26, 2013. Based on a Google Insights analysis, the sample was selected from the top 20 search results for the terms “electronic cigarette,” and “e-cig”. Two searches were performed for each term using the filters “Relevance” and “View Count”. The initial sample included a total of 80 short-length videos from which duplicates were removed, leaving a final sample of 63 unique videos. Data collected included: title; uploader alias; length; number of views, “likes”, “dislikes”, comments and actor demographics. Videos were rated independently by the two reviewers for content theme and connotation. Inter-rater reliability was good (k=.70). Reviewer ratings were compared; consensus was reached on any discrepancies. Data was coded and recorded into an electronic database for statistical analysis. Results: The sample of assessed videos (N=63), generated the following means: video length, 135 seconds; views, 199,311; “likes”, 21.5; “dislikes”, 98.2; and posted comments, 244. Of the actors shown, 49.2% were male, 61.9% were Caucasian, and 33.3%, the largest segment, were coded as young (i.e. < 40 years). The most commonly mentioned brand was Blu. Video content connotation was coded as follows: positive (76.1%); neutral (17.4%); and negative (6.3%). Videos were categorized into one of the following seven content themes: Advertisement (33%); Instructional/how-to (17.4%); news clip/story (19.0%); product review (12.7%); entertainment (11.1%); public health/educational (3.1%); and personal testimonial (3.1%). Conclusions: YouTube videos of electronic cigarettes primarily feature young, male, Caucasian actors and depict e-cigarettes in a positive manner. Most videos were non-traditional advertisements.

P51 Effect of Nicotine on *Streptococcus gordonii* Biofilm Formation and Aggregation. R. HUANG*, M. LI, M. YE, K. YANG, R.L. GREGORY (Indiana University School of Dentistry)

Streptococcus gordonii is a commensal species of human oral flora. It initiates dental biofilm formation and provides binding sites for other colonizers to attach to and generate mature biofilm. Smoking is a risk factor for both dental caries and periodontal disease, and cigarette smoke extract has been reported to facilitate *Porphyromonas gingivalis*-*S. gordonii* biofilm formation. Our hypothesis is nicotine, one of the most important and active components of tobacco, stimulates *S. gordonii* to multiply and generate binding sites. *S. gordonii* planktonic cell growth (turbidity and colony forming units), biofilm formation (crystal violet stain and confocal laser scanning microscopy) and aggregation in PBS with/without nicotine were investigated. Planktonic cell growth was stimulated by 1, 2 and 4 mg/ml nicotine treatment. Biofilm formation was increased by 0.5-4 mg/ml nicotine treatment. CLSM demonstrated bacterial cell numbers were increased in 2 and 4 mg/ml nicotine, but biofilm extracellular polysaccharide was not significantly affected by nicotine. Cell aggregation was upregulated in 4, 8 and 16 mg/ml nicotine with sucrose and in 16 mg/ml nicotine without sucrose. Nicotine stimulates *S. gordonii* planktonic cell growth, biofilm formation and aggregation, which may promote later pathogen attachment to tooth surfaces. (This work was partially funded by the IBASM Student Research Grant (Graduate Level), Indiana University- Purdue University Tobacco Cessation and Biobehavioral Signature Group, and Indiana University School of Dentistry Ph.D. Student Research Fund)

P52 Tobacco Dependence Education in Accredited U.S. Dental Assisting Program Curricula. E.R. SVETANOFF^{1*}, P.T. FORD², C.J. PALENIK², L.M. ROMITO² (¹Butler University, ²Indiana University School of Dentistry)

Tobacco use remains the leading cause of preventable morbidity and mortality in the United States producing numerous adverse oral effects. Thus, tobacco dependence education (TDE) should be an integral part of every oral healthcare professional's education. This study assessed the level of TDE in the curricula of accredited U.S. dental assisting (DA) programs and then compared the findings to those from a similar assessment of dental hygiene (DH) curricula. A previously validated and published 51-item survey was sent to directors of all 298 accredited DA programs. Assessed were curricular TDE content, time spent on each topic, expected levels of clinical competence and resources used. Responses from completed surveys were reviewed, coded and entered into an Excel spreadsheet. Eighty-nine programs returned completed surveys for a 30% response rate. Of the 13 TDE-related content areas, those most often covered were "oral disease related to tobacco use" (100%) and "general diseases related to tobacco use" (93%); those least often covered included "stages of (behavior) change" (29%), "how to develop a comprehensive tobacco intervention program in a private office setting" (23%) and "strategies for community-based tobacco control" (22%). In indicating the level of tobacco cessation intervention (Brief, Moderate or Intensive) students should be able to demonstrate competence in upon graduation. 44% of program directors reported Brief, 54% reported Moderate, while 8% replied Intensive. Less than half of reporting programs conducted a formal assessment of clinical competence in any of the seven TDE-related skills; however, skills in "assessing patient tobacco use" and "associating head and neck findings to tobacco use" were formally or informally assessed by 64% and 61% of respondents, respectively. When compared to DH program survey results, only one of the 13 TDE content areas, "oral diseases related to tobacco use," was covered more often by DA programs. DA programs reported less curricular time for each of the other content areas save "addressing students' own use of tobacco." TDE appears to play a much smaller role in the curricula of accredited U.S. dental assisting programs, when compared to DH programs. DA programs were also less likely to formally assess clinical competence in TDE skills. (Approval was obtained from the IU Institutional Review Board (IRB # 1201007832). Funding: American Dental Education Association Council of Sections 2012 Project Pool)

Clinical Case Reports

DENTAL HYGIENE

CC1 Surgical and Non-Surgical Periodontal Therapy in an Uncontrolled Diabetic. K. BALL*, H. KRAMER, L. MAXWELL (Indiana University School of Dentistry)

Objective: To evaluate healing sites on an uncontrolled diabetic patient following nonsurgical periodontal therapy (NSPT) and osseous surgery in the upper right quadrant. Assessment: A 66 year old Caucasian male patient presented with a chief complaint of wanting a cleaning. The patient reported no dental cleanings for 2 years. His medical history was extensive including diabetes that was uncontrolled. His A1C ranged from 7.5 to 9.6 during the course of treatment. The patient stated that he brushed twice a day and flossed 3-5 times a week. The clinical examination of the UR quadrant revealed generalized chronic periodontal disease with probing depths ranging from 2-6mm and clinical attachment levels ranging from 2-7mm. The patient also presented with generalized moderate plaque-induced gingivitis. The intraoral radiographs revealed generalized mild bone loss in the upper right quadrant. DH Care Plan: The initial treatment planned for the patient was four quadrants of (NSPT), extensive oral hygiene instructions (OHI), and a periodontal tissue re-evaluation. Evaluation: The periodontal re-evaluation from (NSPT) showed improved probing depths of 1-2mm on several teeth, while the other teeth showed no improvement at all. The re-evaluation presented with no improvement in the health of the gingival tissue and a bleeding index of 16. During the consultation with the dentist, it was determined that the patient should return for periodontal maintenance in 3 months. At the 3 month recall, periodontal probing comparison revealed a 2mm increase in probing depths on several teeth. The dentist then referred the patient to the Graduate Periodontics Department for further evaluation. Surgical therapy was recommended in the upper right and lower right quadrants. Follow-up: The upper right quadrant was treated by a Periodontal Resident and a tissue re-evaluation was completed. The surgical therapy resulted in 1-3 mm improvement in probing depths on sites with initial probing depths of 4+mm in the upper right quadrant. The patients gingival health demonstrated improvement as measured by reduced bleeding, improved color and contour, and a lowered plaque score. Conclusion: Research indicates that patients with uncontrolled diabetes have an increased risk for periodontal disease and poor wound healing capability; however this patient was able to see improvement of periodontal health at the three month mark despite his uncontrolled diabetes. This case is ongoing.

CC2 Can Patient Compliance Improve Gingiva and Periodontal Status. M. NGUYEN*, C. SCHAFER, H. YATES, P. RETTIG (Indiana University School of Dentistry)

Objective: The objective of this clinical case presentation is to evaluate the correlation of oral hygiene with gingival and periodontal improvement after scaling and root planning, and to educate dental providers and patients on the importance of patient compliance with professional recommendations in order to achieve optimum results from treatment. Background: A thirty-six year old patient present with a negative medical history. The patient had not received any dental treatment in the past five years. The patient's last dental hygiene treatment was an adult prophylaxis. Clinical examination: Clinical findings for this patient included generalized moderate chronic periodontist as evidenced by 4-6 mm CAL with localized severe chronic periodontist as evidenced by 8 mm CAL on # 18. The patient presented with a bleeding index of 21%, an O'Leary plaque score of 64%, and generalized moderate to heavy subgingival and supragingival calculus. Additional findings included generalized 1-2 mm of gingival recession and localized incipient furcation involvement on #18. Radiographically, this patient presented with generalized mild bone loss as evidenced by 3-4 mm measurement from the CEJ to the alveolar crest and localized moderate bone loss as evidenced by 5 mm measurement from the CEJ to the alveolar crest on tooth #

28. Patient also presented with generalized moderate plaque induced gingivitis as evidenced by red, rolled, and spongy gingiva. Treatment: The treatment plan consisted of four quadrants of scaling and roots planing therapy, dental health education and a tissue re-evaluation. Results: Following treatment, the tissue re-evaluation was performed at the patient's 3 month recall due to scheduling constraints. The patient presented a positive tissue response with generalized improvement in clinical attachment levels (CAL) as evidenced by generalized 4-5mm CAL and localized 4-6 mm CAL in the molars. The gingival tissue displayed a decrease in redness and appeared less rolled at the margins. The patient's bleeding index decreased by 6% and their O'Leary plaque score decreased by 22%. Conclusion: The positive response is the result of hygienist's thorough scaling and root planning therapy, extensive patient education and patient compliance to oral hygiene recommendations.

CC3 Change in Dental Health Values and Beliefs Through Education. J. ALLEN*, N. DODGEN, H. TAYLOR (Department of Dental Hygiene, Indiana University School of Dentistry, Indianapolis, Indiana)

Objective: The objective of this clinical case report is to evaluate the change in dental health values and behavioral habits in a patient with poor periodontal health and high caries risk. Assessment: A 38 year-old female, Caucasian patient reported to the dental hygiene clinic with the chief complaint of "I want to keep as many teeth as possible". The patient reported no dental or health care in over twenty years. Patient's medical history is negative. The patient presented periodontally with generalized moderate, plaque induced, diffuse gingivitis as evidenced by red, bulbous, soft gingiva with generalized subgingival and supragingival calculus. Patient also had a bleeding index of 15%. In addition, generalized healthy periodontium with localized mild alveolar bone loss. Patient presented to the dental hygiene clinic with localized chronic periodontitis and severe local periodontitis. A caries risk assessment led to the following findings: no dental home for over twenty years, active caries, past history of daily intake of soda, generalized plaque, retained root tips, and radiographic periapical pathology. The patient also reported that she had never used floss. In addition, the patient reported initially with a plaque score of 62%. Dental Hygiene Care Plan: Three quadrants of scaling and root planing with a tissue evaluation in six to eight weeks, shortened recall intervals, extensive oral hygiene instruction (OHI), education to evaluate clinical signs of disease at home and diet analysis/counseling were planned. Patient will also be referred for restorative needs. Treatment: Completed oral hygiene instructions, diet analysis/counseling, three quadrants of SRP, and tissue evaluation six to eight weeks post treatment. Evaluation: After education and initial non-surgical periodontal therapy, this patient showed clinical improvement with regards to gingival status and a decrease in periodontal probing depths (1-2mm generalized improvement). In addition to clinical dental improvements, the patient's behavioral habits have changed dramatically as evident by compliance to OHI and diet counseling and a reduced plaque score of 22%. The patient's post treatment appointment revealed improved gingival status from diffuse gingivitis to marginal/papillary gingivitis and 1-2mm generalized improvement in probing depths. The patient also had a 3% reduction in her bleeding index. Educating a patient on proper diet, clinical signs of disease and treatment can result in positive changes to the patient's dental health values and behavioral habits.

CC4 Non-surgical Therapy on Chronic Periodontitis with Pseudo-pocketing from Calcified Deposits. S. MADSEN*, M. GRIMMER, A. RIECK (Indiana University School of Dentistry)

Objective: The objective of this clinical case presentation is to assess the outcomes of nonsurgical periodontal therapy by a dental hygiene student on a patient with generalized chronic periodontitis with pseudo-pocketing from calcified deposits. Assessment: A 26 year old Asian male presented to the dental hygiene clinic wanting his "teeth cleaned". Patient presented to clinic with several fractured teeth, abscesses, and defective restorations. The patient reported having dental work out of the country, but

had not had a cleaning in a long time. Radiographically, patient presented with generalized mild bone loss as evidenced by bone levels being 3-4mm from the CEJ to the crest of alveolar bone. There was localized moderate bone loss as evidenced by bone levels being 5-6mm from the CEJ to the crest of alveolar bone on tooth numbers: 30 Distal, 22-27 Mesial and distal, 19 Mesial, and 21 Mesial. Clinically, patient presented with clinical attachment levels from 4-7mm. Patient presented with some pseudo pocketing on areas with increased probing depths. Patient also presented with generalized moderate gingivitis as evidenced by dark red, bulbous, spongy gingiva, and significant bleeding on probing in all areas. DH Care Plan: Four quadrants of scaling and root planning, local anesthesia for the vasoconstrictor to reduce bleeding, oral hygiene instruction, and tissue re-evaluation were completed. Patient was referred to Comp care to treat abscesses. Evaluation: At the tissue reevaluation patient presented generalized better probing depths and less negative recession on the anterior teeth. Patient showed no improvement in probing depths in the anterior teeth. Gingivally, patient presented with better color and contour and slightly less bleeding. Patient showed slight improvement and is going to continue treatment in the dental hygiene clinic on a three month recall to improve periodontal health.

CC5 Non-Surgical Periodontal Therapy on an Uncontrolled Type I Diabetic Patient. K. YESNIK*, T. TRAUB, N.A. YOUNG (Indiana University School of Dentistry)

Objective: The purpose of this clinical case report was to evaluate the outcomes of non-surgical periodontal therapy on an uncontrolled Type I diabetic patient. Assessment: A 53 year-old Caucasian male presented to the Dental Hygiene clinic with uncontrolled Type I diabetes and a history of smoking cigarettes. Due to his battle with smoking cessation, he gained 40 pounds in one year in an attempt to manage his nicotine withdrawals. Increased frequency of consuming fermentable carbohydrates lead to higher A1C hemoglobin levels. Oral hygiene had also worsened during this period. A clinical intraoral examination revealed generalized, moderate, plaque-induced gingivitis and generalized periodontitis modified by systemic factors, i.e. uncontrolled Type I diabetes and smoking. DH Care Plan: Scaling and root planing (SRP), periodontal re-evaluation, and periodic dental examination were planned. Oral hygiene instruction included a discussion on the link between diabetes, smoking, and periodontal disease and the importance of a healthy diet. Treatment: During the Fall of 2013, the DH care plan was implemented. Evaluation: Treatment results indicated a generalized improvement in gingival and periodontal health with reduced probing depths and no bleeding. The patient was referred to the Graduate Periodontics department for surgical treatment on localized areas of bone loss that did not respond to non-surgical periodontal therapy. Conclusion: Complications such as uncontrolled diabetes and continued smoking make it difficult to achieve total health when treating periodontal patients and offer challenges in terms of continued patient motivation. Patients with these problems are likely to require referral to the periodontist.

CC6 Pregnancy Induced Gingivitis in an Adolescent Patient. N. MOTE*, M. MULLINS, P. RETTIG (Indiana University School of Dentistry)

Objective: The objective of this clinical case presentation is to educate dental health providers on the management of an adolescent patient that presents with pregnancy gingivitis. Background: A seventeen year old pregnant patient presented for a prophylaxis. The patient had a history of inadequate oral hygiene and had not received regular dental care since 2010. A daily diet included high amounts of fermentable carbohydrates that contributed to a high caries risk status. Additionally, the patient had been treated for a history of alcoholism in 2012. Clinical examination: Patient presented with generalized moderate, plaque induced gingivitis modified by pregnancy. This was evidenced by shiny, red, edematous gingival tissue with rolled margins, bulbous papilla and loss of tissue resiliency. The periodontal description revealed generalized bleeding on probing and multiple areas of pseudo-pocketing

evidenced by 4mm probing depths with 1mm of negative recession. A plaque score was taken and disclosed that 32% of the patient's tooth surfaces contained plaque. Treatment: The treatment plan for therapy consisted of a routine prophylaxis and dental exam. Oral health instruction was provided to both the patient and guardian. Information and resources regarding both pregnancy and alcohol abuse were also made available to the patient. An application sodium fluoride varnish was provided to prevent the development of caries. Results: The patient tolerated treatment well. Instrumentation with ultrasonic instruments was provided to help reduce inflammation and irrigate bacteria in the sulci. Following instruction, the patient was able to demonstrate proper Bass brushing and c-shape flossing techniques along with the use of a floss aid in the 3rd molar region. After oral health education the patient showed an understanding of the pregnant body's response to plaque and importance of maintaining proper oral hygiene, as well as, a healthy diet. A dental exam revealed no need for restorative treatment or referral. In a follow up the patient reported improved oral hygiene and dietary habits. Furthermore, the patient stated that she had recently joined a new support group for alcohol addiction. Conclusion: Maintaining a healthy oral cavity during pregnancy can be achieved through extensive oral health education, prophylaxis and patient compliance. Plaque induced gingivitis modified by pregnancy can be reversed with proper treatment and regular care. Factors such as, early intervention, patient education, support from family and healthcare providers can have a positive impact on oral and overall health. These type of interventions also contribute to the health of the unborn child.

CC7 Periodontal Tissue Response by Periodontal Debridement, Chlorhexidine, and Antibiotics.
J. SPENCER*, D. SPERRING, R.H. RACKLEY (Indiana University School of Dentistry)

Objective: The objective of this clinical case presentation is to evaluate the outcomes of periodontal tissues by treatment with Chlorhexidine, Antibiotics and Periodontal Debridement on a patient with severe gingivitis. Background: A 19 year-old female presented with a chief complaint of "my wisdom teeth hurt". The patient reported no dental care since she was 9 years old. Her medical history was essentially negative other than current birth control regime after recent child birth. The patient stated that she sometimes brushes daily and rarely flosses. Assessment: At the first appointment the patient presented with generalized, severe, plaque-induced gingivitis as evidenced by deep red, bulbous, and loose gingival tissues. Due to the extent of severe gingivitis, the patient was prescribed 500mg of Amoxicillin to take 4 times a day for one week in addition to Chlorhexidine Gluconate rinse to use twice daily. Patient presented with 2-4mm CAL with the 4mm pockets due to pseudo-pocketing caused by severe inflammation. Dental Hygiene Treatment Plan: Adult Prophy with extensive oral hygiene instruction. Treatment: The treatment was achieved throughout four appointments where antibiotics and Chlorhexidine were prescribed, and periodontal debridement was completed. Results: A week after the initial visit the patient's gingival description improved the most in the posterior regions and slightly in the anterior regions due to compliance with Chlorhexidine and Antibiotics. At the next two appointments, the patient's gingival description continued to improve slightly. At the final appointment when the treatment was completed, the patient's gingival description had significantly improved. Conclusion: Antibiotics, Chlorhexidine, and Periodontal Debridement are successful in treating severe gingivitis.

CC8 Non-Surgical Periodontal Therapy on a Periodontal Patient. V. BUNTON*, E. HAMILL, L. MAXWELL (Indiana University School of Dentistry)

Objective: The objective of this clinical case study is to evaluate the outcomes of non-surgical periodontal therapy by a dental hygiene student on a patient with chronic periodontitis. Background: 49 year old Hispanic male reported on 6/20/13 with concern about the health of his gingiva around the lower anterior teeth. The patient reported that he had never had a dental cleaning before. The patient stated that he brushed twice a day, but seldom flossed. His medical history revealed presence of type II diabetes

mellitus, hypertension, and hyperlipidemia. All conditions are currently controlled with diet and medication. Clinical Examination: The clinical examination revealed generalized severe plaque-induced diffuse gingivitis evidenced by dark red color, bulbous contour, and spongy consistency. The patient also presented with generalized chronic periodontal disease with probing depths ranging from by 2-7 mm and clinical attachment levels ranging from 2-8mm. The intraoral radiographs revealed generalized moderate bone loss. Treatment by Dental Hygiene Student: Prescription Peridex was advised and given to the patient. Four quadrants of scaling and root planing (NSPT) were completed. Extensive oral hygiene instructions were given. Tissue re-evaluation was completed, and the patient was referred to Graduate Periodontics department for comprehensive evaluation. Results by Dental Hygiene Student: Scaling and root planing resulted in overall improvement. There was an improvement in bleeding scores, probing depths and clinical attachment levels. Even though the plaque score improved from 83% to 45%, the patient's oral hygiene may not have been a factor in his improved gingival health. The patient was given extensive oral hygiene instructions at each visit, but his plaque score did not improve significantly over the course of several appointments. Conclusion: Research indicates that non-surgical periodontal therapy, alone, is effective regardless of the patient's homecare. The patient had significant healing and improvement without significant improvement of his oral homecare; but for how long? This case is ongoing.

CC9 Effects of Poorly Controlled Diabetes on the Progression of Periodontitis. B. HOPF*, R. FRANKER, J. BLANCHARD (Indiana University School of Dentistry)

The objective of this case presentation is to discuss the association of poorly controlled diabetes and poor oral hygiene on the progression of periodontitis. Assessment: A 54 year old Caucasian male presents to the Dental Hygiene Clinic for a compromised periodontal maintenance with a chief complaint of tooth mobility and concern for losing teeth. The medical history reveals a history of hypertension, hyperlipidemia, diabetes mellitus type 2, coronary artery bypass graft, and Bell's palsy. During the course of treatment he reports fasting blood glucose levels >300mg/dl and HbA1c of 10%. He reports brushing twice a day and rarely flosses. He also expressed difficulty with brushing due to a hyperactive gag reflex and tooth mobility. Clinical Assessment: In 2007 this patient presents with moderate periodontitis as evidenced by clinical attachment levels (CAL) of 5-7 mm. This patient was then seen in 2013 and presented with generalized moderate to severe periodontitis as evidenced by radiographic bone levels of 5-10 mm as measured from crest of the alveolar bone to the CEJ and clinical attachment levels (CAL) of 5-11 mm. The patient also presents with generalized severe marginal and papillary gingival inflammation. Dental Hygiene Care Plan: A new medical consult was sent to his physician to understand the nature of his diabetic condition. The medical consult was returned which stated, "fasting blood sugars are tested by the patient each day: low 259, high 400. Patient is adherent with medication regimen. Glycemic control has been poor. The patient reports no symptomatic hypoglycemic episodes. The patient is watching his diet." Patient received oral hygiene instruction, four quadrants of scaling and root planing (SRP) with the use of local anesthesia (2% Lidocaine with 1:100,000 epinephrine) and periodontal tissue reevaluation six weeks after SRP. Evaluation: Although there was a slight improvement in gingival inflammation following dental hygiene therapy, clinical attachment levels were unchanged. Oral hygiene also showed no improvement. Conclusion: Unfortunately, the patient's poor glycemic control and lack of good oral hygiene make him a poor candidate for further periodontal therapy unless blood glucose levels and plaque control improves. He was referred to Graduate Prosthodontics and Graduate Periodontics for further evaluation and treatment.

CC10 Microstomia Management of Patients with Sjogren's Syndrome Secondary to Scleroderma.
J. FORRESTER, B. POWERS*, L. MAXWELL (Indiana University School of Dentistry)

Objective: The objective of this case presentation is to evaluate the outcome of microstomia management of patients with Sjogren's syndrome secondary to Scleroderma. Assessment: A 60 year-old female presented with Sjogren's syndrome secondary to a previous diagnosis of Scleroderma. The patient presented with a history of high caries risk, DMFT score of 19, plaque score of 38%, and generalized moderate supragingival and subgingival calculus. Oral facial involvement of the disease in this patient has greatly compromised her ability to open her oral cavity. It was established that the patient experiences extreme difficulties with oral hygiene self-care due to the constriction of her oral tissue. DH Care Plan: The patient's difficulty of care level was established as a dental hygiene class III. This level was determined based upon her caries risk assessment, medical history, calculus accumulation, and overall plaque score. Self-care instructions were planned and presented to enhance her oral home care routine. The supplemental recommendations are as follows; to improve dexterity while brushing, switch from an adult standard manual toothbrush to a pediatric sized power toothbrush. This change will enhance caries prevention and further destruction to the periodontium by more effectively reducing plaque accumulation and ultimate calculus formation. She was encouraged to switch from an alcohol based oral mouth rinse to a non-alcohol based oral mouth rinse to lessen the symptoms of dry mouth. Lastly, extensive education regarding Sjogren's syndrome, as it relates to the development of dental caries and periodontal disease, was also presented to the patient. Evaluation: This patient was seen six months after the implementation of the dental hygiene care plan. At that time, she reported improvement in oral home-care due to enhanced manual dexterity provided by the use of the pediatric power brush. This was evidenced by a 60% reduction in plaque score. She also noted intermittent relief of dry mouth, a side effect of Sjogren's syndrome, after switching to a non-alcohol based oral mouth rinse. Conclusion: The result of this case presentation demonstrates how patient education combined with the incorporation of adaptive accommodations such as toothbrush type, power vs. manual, toothbrush size and choice of oral mouth rinse has effective results in the reduction of the adverse oral manifestations that can occur in patients with microstomia as a result of Sjogren's syndrome secondary to Scleroderma.

CC11 Poor Oral Health and Diet: What Part Does Socioeconomics Play? L. HUFFMAN*, N. KELLAR, L. MAXWELL (Indiana University School of Dentistry)

Objective: The objective of this case report is to demonstrate how much socioeconomics affects dental access and oral health. Assessment: A 24 year old Caucasian male who has never received dental care presented to our clinic. His medical history indicates a history of testicular cancer and the patient has smoked for three pack years. The patient presented with severe - moderate diffuse gingivitis with heavy BOP. The patient also presented with 4-8 mm of CAL but with healthy bone levels 1-2 mm from the CEJ radiographically. Heavy subgingival and supragingival calculus was detected on all teeth along with heavy plaque collection on all posterior teeth along with active decay. DH Care Plan: A full mouth debridement with local anesthetic was performed and a 0.2% Chlorhexidine rinse to reduce active gingivitis was given. Dietary analysis was obtained, OHI given, and a re-evaluation was performed one week later. When the patient returned it was determined that an adult prophylaxis would be sufficient treatment along with extensive oral hygiene instructions. Treatment: An adult prophylaxis was performed on the patient with the use of Oraquix for pain control. Oral hygiene instructions were given to the patient with focus on the patient's posterior teeth where plaque control was an issue. The patient also received nutritional counseling. The patient was asked to return in one month for a re-evaluation. Evaluation: After a periodontal re-assessment it was determined that the treatment had been effective, in that attachments and probing depths improved by 1-3mm overall. There was a significant reduction in bleeding and gingival color and consistency had greatly improved. The patient's daily oral hygiene also improved

significantly. The patient was still a high caries risk due to his active decay, but no new decay was noted at his return. Conclusion: Patient education is crucial for a patient with a history of no dental care. Studies show that poor childhood and adult socioeconomic status, when experienced by the same individual, lead to poor dental health. Studies also show that individuals with low income have poorer diets that are higher in fat composition with fewer nutritional sources. Dental hygienists are in a unique position to counsel their patients on proper nutritional intake and aiding their patients in furthering their overall oral health.

CC12 Modification of Dental Hygiene Care Plan in a Dementia Patient. K. LAMPING, J. JANOSKI*, A. REED (Indiana University School of Dentistry)

Objective: The objective of this clinical case presentation is to establish effective communication between the patient with dementia, as well as the caregiver, and identify necessary modifications throughout the dental hygiene appointment. Assessment: An 81-year old, Caucasian male patient presented with caregiver who stated the chief complaint of “my husband needs a cleaning.” The patient’s wife explained that his last cleaning was over 24 years ago. The patient presents with dementia, Type II Diabetes, hypertension, prostate enlargement and cardiovascular disease, in addition to polypharmacy. Because of the patient’s mental health disorder, the patient is unable to adequately communicate with the clinician, so the caregiver is present at all times. The caregiver stated that the patient brushes at least once per day, never flosses but rinses with Listerine everyday. The patient’s Type II Diabetes is controlled with a healthy, low carbohydrate diet. This is especially important, not only for the diabetes control, but also because of his high risk for dental caries. The patient is at high risk for dental caries due to active decay, no dental home, medication-induced xerostomia and inadequate home care due to mental health disorder. He presented clinically with generalized moderate plaque-induced, diffuse gingivitis as evidenced by light pink, soft, rolled gingiva with moderate bleeding on probing and generalized chronic periodontitis as evidenced by 4-6 mm clinical attachment levels. Radiographically, the patient presented with generalized mild horizontal bone loss as evidenced by 3-4 mm measured from the cemento-enamel junction to the crest of the alveolar bone. DH Care Plan: Four quadrants of scaling and root planing with extensive oral hygiene instructions, and periodontal tissue re-evaluation were performed. Evaluation: Upon re-evaluation, no improvement was seen with the patient’s oral hygiene or periodontal status. The dental hygiene care plan was adapted by implicating aggressive prevention, such as shorter recall intervals for periodontal maintenance, application of professional topical fluoride and extensive oral hygiene instruction modified towards the caregiver. Multiple auxiliary aids were introduced to the caregiver, specific to the patient’s needs. Verbal and nonverbal communication techniques were utilized. Conclusion: Concluding this clinical case, it was apparent that many modifications must be made when managing a patient with dementia. When the patient is unable to perform basic oral hygiene care on his or her own, the responsibility of home care lies in the hands of the caregiver. Hence, the focus of oral hygiene instruction becomes more about educating the caregiver.

CC13 Preventing Caries and Periodontal Disease on a High Risk Patient. A. MURPHY*, A. CARLTON, P. RETTIG (Indiana University School of Dentistry)

Objective: The objective of this clinical case presentation is the implementation and evaluation of preventive measures by a dental hygiene student on a patient who presents with high caries risk and localized periodontitis. Background: A 54 year old Caucasian male patient presented as a 3 month prophylaxis recall with a chief complaint of “sensitivity in one tooth in the upper right quadrant.” His medical history was positive for hyperlipidemia, hypertension, diabetes mellitus type II, prior head and neck radiation, and anxiety and depression. He takes multiple medications which may induce xerostomia, taste disorder, and gingival hyperplasia. The patient presented with generalized healthy radiographic bone levels measuring 1-2mm from the CEJ to the alveolar crest. The patient presented with generalized

moderate plaque-induced marginal and papillary gingivitis as evidenced by red, bulbous and rolled gingiva which is spongy in consistency. The patient also presented with generalized healthy periodontium as evidenced by 1-3mm CAL, with localized areas of 4-6mm CAL. The patient's plaque score at the last recall visit was 28%, which was an improvement by 36% from our first appointment. The patient presents with numerous modifying factors and a fair adherence to protective modifying factors. DH Care Plan: Patient will be placed on a 3 month recall interval as an adult prophylaxis patient, which includes debridement, oral hygiene review, nutritional counseling, 5% NaF varnish application, and evaluation for the need of prescription fluoride. Evaluation: The patient's plaque score and gingival health have continually improved over his course of treatments in the dental hygiene clinic. The patient was receptive to the information provided during his dietary analysis session, and he understands the correlation between his systemic and oral health. Probing depths and areas of demineralization are not progressing at this time. Conclusions: Positive results have occurred from the preventive treatment plan. It is critical to continue monitoring the patient's systemic health, caries risk, and periodontal conditions to evaluate the long-term success of the preventive measures applied, but short-term results have shown continued improvement.

CC14 Contrasting Attitudinal Differences in Oral Care Among Two Countries. A. LENNON, K. HERTER*, L. MAXWELL (Indiana University School of Dentistry)

Objective: To evaluate the attitudinal differences toward oral care in the United States versus India. Background: Patient relocated from India to the United States seven years ago stating that in India dental health is not promoted, nor is it a priority. The patient reported never having a dental home, prophylaxis or oral hygiene instruction. Assessment: A 64 year old Indian female patient presents with chief complaint of "Why do my teeth look different from others?" Her medical history was essentially negative except for asthma. The patient stated that she brushed twice a day with a manual toothbrush, had never flossed and had never heard of fluoride. Clinical examination revealed chronic periodontal disease with probing depths and clinical attachment loss ranging from 2-7mm. Patient also presented with generalized moderate marginal plaque-induced gingivitis as evidenced by dark pink, firm, rolled margins and blunted papilla with moderate BOP. Patient also presented with localized severe marginal plaque-induced gingivitis on the lower anteriors as evidenced by red, rolled margins, bulbous gingiva, blunted papilla and moderate BOP. Patient's plaque score recorded was 71%. Intraoral radiographs revealed generalized mild bone loss as evidenced by 3-4mm horizontal bone loss and 5mm moderate bone loss localized on tooth numbers 1D, 6D, 15D, 18M and mesial of 22 through the distal of 27. DH Care Plan: Four quadrants of scaling and root planing (SRP) with local anesthetic, extensive oral hygiene instructions (OHI) and a periodontal tissue reevaluation were completed. Evaluation: Periodontal reevaluation revealed 1-2mm improvements in some areas with some localized advancement of probing depths. Patient was referred to Graduate Periodontics for further evaluation. The SRP resulted in significant improvement in bleeding however with minimal changes in probing depths. Patient's home care improved greatly which reflected significantly healthier gingival tissue as evidenced by pink, firm, slightly rolled margins and slightly blunted papilla. Over the course of treatment and repeated OHI, patient's PS was reduced to 13% generalized. Conclusion: Although additional periodontal treatment is needed, the patient's knowledge concerning oral health has significantly increased as well as her attitude and understanding of adequate home care with regular dental visits.

CC15 Staining Potential of Cetylpyridinium Chloride - A Literature Review. L. GEBREMICHAEL*, C. STEVENSON, S. PHILLIPS (Indiana University School of Dentistry)

The purpose of our literature review was to attempt to identify a link between extrinsic staining similar to the type of stain we have witnessed in our patients, and certain brands of oral rinses. A literature review

provides evidence that cetylpyridinium chloride (CPC) may be responsible for extrinsic staining in certain individuals. CPC is the main ingredient in some over the counter mouth rinses. One study we reviewed identified and described the active ingredient in various mouth rinses that causes extrinsic stain. One objective of our review was to determine whether the absence of water can influence the amount of stain that occurs when mouth rinses containing (CPC) are used. Another objective was to discuss whether saliva and/or tea can influence the presence of extrinsic staining in patients using a (CPC) rinse. The staining potentials of CPC were demonstrated by a study where specimens were soaked in human saliva, and washed in distilled water, then immersed in various CPC containing mouth rinses. After immersion of the specimens in CPC containing mouth rinses, the specimens were washed again with distilled water then exposed to 10 ml of standard tea solutions. The second study performed involved blocks that were immersed into either human saliva or water, then they were rinsed with distilled water, then placed into a CPC containing mouth rinse, rinsed again with distilled water, then placed into a tea solution for one hour. The results of our literature review suggest that (CPC) containing mouth rinses may cause extrinsic stain in some individuals. Our review also suggests that the presence of water can decrease the amount of stain and the presence of saliva and/or tea can increase the amount of stain that appears in relation to cetylpyridinium chloride (CPC).

CC16 Non-surgical Periodontal Treatment Prior to Transplant Surgery. K. LUNDQUIST*, T. GEBHART, P. RETTIG (Indiana University School of Dentistry)

Objective: To educate the dental professional regarding a patient with an extensive medical history and limited previous dental care. Background: A 55 year old female new patient presents to the dental hygiene clinic with an extensive medical history which includes: Asthma, chronic obstructive pulmonary disease, diabetes type II, hypothyroidism, seasonal allergies, sleep apnea and history of cancer. The patient is currently a candidate for a lung transplant. Patient reports tobacco use until August of 2013. Patient had not accessed dental care since 2009. Assessment: Patient presents with generalized severe plaque induced and non-plaque induced, diffuse gingivitis as evidenced by dark pink to red, soft, and bulbous gingiva. Patient also presents with generalized mild periodontitis as evidence by 4-5 mm CAL. Radiographically patient presents with localized mild periodontitis as evidence by 1-2 mm of bone loss with localized moderate periodontitis as evidence by 3-4 mm of bone loss. Patient's oral hygiene status included generalized heavy plaque ranging from 32-61% and high caries risk due to carious lesions present. Dental Hygiene Care Plan: Patient received scaling and root planing in all four quadrants with local anesthesia. She also was given extensive oral hygiene instruction. Patient has not returned for tissue re-evaluation at this time. Evaluation: Over a six week time period the patient's gingiva improved as evidence by more light pink/less red, more firm, and less loose tissue. Conclusions: Treatment was successful as evidence by tissue improvement, less bleeding, and patient report of decreased sensitivity and pain of her teeth.

CC17 Side Effects of "Miracles": Treatment of Patient with Osteonecrosis. R. SHRESTHA*, S. HOLLOWELL, R.H. RACKLEY (Indiana University School of Dentistry)

Objective: The objective of this clinical case presentation is to educate dental care providers about the modifications required for dental treatment on a patient with bisphosphonate related osteonecrosis of the jaw (BRONJ). Assessment: A 76 year old Caucasian female patient presented for a prophylaxis and exam with a complaint of sensitivity in the lower right quadrant of mouth. Her medical history was positive for hyperlipidemia, gastroesophageal reflux disease, hypertension, osteoporosis, a colonoscopy, and eyelid surgery. The patient stated that she brushed twice a day and flosses daily. Clinical examination revealed localized, chronic periodontitis on teeth #3 and 19 as evidenced by 4-5mm CAL. Patient also presented with generalized, mild, plaque-induced gingivitis. The intraoral radiographs revealed localized

mild bone loss on teeth #s: 11D, 18M, 19D, 22D. DH Care Plan: Patient's difficulty level was classified as a class II prophylaxis. Treatment: Entire dentition was debrided and OHI was given. A dental exam was performed and it was determined that the area of osteonecrosis was stable. Evaluation: Although modification was required in the lower right quadrant due to the presence of sensitivity, patient tolerated treatment well. Conclusion: Though side effects to drugs like osteonecrosis are rare, they are still present in society. Therefore, it is important that dental care providers be knowledgeable about the effects of these drugs and how to treat these cases because a negative side effect can be a precursor of a malignancy or an even more serious problem.

CC18 Case Presentation of a Patient with Bell's Palsy. A. GARDNER*, S. PARKER, P. RETTIG
(Indiana University School of Dentistry)

Objective: The objective of this clinical case presentation is to demonstrate the impact of Bell's Palsy on a periodontal maintenance patient. Assessment: A 92 year old male presented accompanied by a caregiver with a chief complaint of "I would like to get my teeth cleaned." The patient had not received dental care in over a year. His medical history included a history of angina, artificial joint, atherosclerosis, Bell's palsy, skin cancer, heart murmur, hyperlipidemia, hypertension, pacemaker, rheumatoid arthritis in the back and shoulders, blindness, and in a wheelchair. The patient reported that he brushed two times a day and flossed several times a week. Clinical examination revealed missing teeth 1, 4, 5, 13, 14, 16, 17, 21, and 30-32. Patient presented with chronic periodontitis and a generalized clinical attachment level of 4-5mm and localized 8mm on #19 with furcation involvement. Patient also presented with generalized healthy gingiva and localized mild gingivitis around the molars. Radiographs revealed generalized mild bone loss with evidence of furcation involvement on teeth 2, 3, and 19. DH Care Plan: periodontal maintenance, extensive oral hygiene instruction (OHI), and fluoride application were completed. Evaluation: Patient's periodontium will be evaluated at the next 3 month recall visit and assessment will be conducted for the possibility of undergoing scaling and root planing again. Conclusion: With proper home care and maintaining 3 month recall intervals, the patient should remain in a stable periodontal status.

CC19 Oral Health Needs of Dementia Patients in Assisted Living Homes. K. FLOYD*, A. KIMBRELL, L. MAXWELL (Indiana University School of Dentistry)

Objective: To evaluate the oral care of a dementia patient in an assisted living home. Assessment: A 92 year old Caucasian female patient presented with the chief complaint of "getting my teeth cleaned". Her medical history was positive for coronary artery disease, hypertension, dementia and hallucinations. The patient stated that she brushed "whenever I go to the bathroom" and rarely flossed. Clinical examination revealed extensive dental needs including multiple retained root tips and decay. She had a moderate plaque score of 18% with material alba found under crowns where recurrent decay was present. Intraoral radiographs revealed multiple teeth with recurrent decay. Patient did not wish to be referred to comprehensive care for treatment of caries due to her "old age" and not being in any pain. DH Care Plan: Prophy was completed. Extensive oral hygiene instructions were given verbally to patient and caregiver. Oral hygiene instructions were also written down for assisted living home staff. Patient placed on 3 month recall due to inadequate home care. Follow-up: Patient returned to DH Clinic for 3 month recall visit. Assessment revealed home care had worsened and plaque score increased to 46%. Additionally, a crown had fractured off at the gumline since her last visit. Evaluation: While extensive oral hygiene instructions were given to the patient, her mental status had an effect on her ability to retain and implement the oral hygiene care instructions. Conclusion: It is essential that the staff at assisted living homes receive training from dental hygienists on how to provide proper oral hygiene care to their residents as well as assist patients in caring for their teeth.

ENDODONTICS

CC20 Surgical Management of a Large Periapical Cyst. B. PRATHER*, Y. EHRLICH, K. SPOLNIK (Indiana University School of Dentistry)

Objective: This case report presents the surgical endodontic treatment of a large periapical cyst and the management of a medically compromised patient. Methods: A 65 year old male with a medical history significant for hypertension, hyperlipidemia, type 2 insulin dependent diabetes mellitus, post traumatic stress disorder, gastroesophageal reflux disorder, and obstructive sleep apnea presented with a chief complaint of pain to palpation in the vestibule over teeth 7 and 8. Radiographic exam showed a 2 x 2 cm, unilocular radiolucency centered over the apex of #7 and extending over the apex of #8. #7 had previous endodontic treatment with a post and core, and the pulp of #8 tested vital. Both were restored with sound porcelain fused to metal (PFM) crowns, present for over ten years. The size of the lesion and the tendency of odontogenic keratocysts to occur in this area in older individuals, along with patient preference to keep his natural dentition and current restorations indicated surgical removal and histopathology examination. Treatment: Intentional non-surgical root canal treatment was completed on tooth #8 due to probable devitalization during surgery. Surgical techniques consisted of reflection of a flap to reveal a cavity filled with a milky-white fluid, curettage of the cyst lining, root preparation with a surgical ultrasonic tip and Brasseler's Endodontic Root Repair Material putty. Emdogain, demineralized freeze dried bone allograft and Dynamatrix membrane were used to graft the residual bony crypt. Management of the patient's medical problems included stress reduction protocol, blood pressure and glycemic control measurements, and limitation of epinephrine. Results: A histologic diagnosis of periapical cyst was given. Initial follow-up visits showed resolution of symptoms and uneventful soft tissue healing. Conclusion: Successful treatment of a large periapical cyst can be achieved without disassembly of coronal restorations through surgical endodontic therapy in a medically compromised patient.

CC21 Decoronation of an Ankylosed Tooth: A Case Report. A.Z. NERNESS*, A.C. TROXEL, K.J. SPOLNIK, Y. EHRLICH (Indiana University School of Dentistry)

Ankylosis is a potential negative side effect of dental trauma and can lead to infraocclusion and disruption of normal dentoalveolar development in growing individuals. The decoronation procedure can be used to remove an ankylosed crown thus preserving bone and permitting normal bone growth and development. In this case report, an ankylosed tooth is treated with the decoronation procedure. A patient with a history of severe facial trauma from a horse kick presented with infraocclusion and ankylosis of tooth #29. A decoronation procedure was performed and the patient was placed on standard trauma recall regimen. Results: Following the procedure, healing was successful and the alveolar ridge height and width was maintained. This case demonstrated decoronation as a viable treatment option for ankylosed teeth in growing individuals.

CC22 Management of Spontaneous Anti-resorptive Osteonecrosis of the Jaw Associated with a Non-Bisphosphonate Drug. K.W. KIM*, J.C. JACOBS, A. GHONEIMA, A.Z. NERNESS, K.J. SPOLNIK, Y. EHRLICH (Indiana University School of Dentistry)

Objective: To report a case of osteonecrosis of the jaw (ONJ) in a patient on a new non-bisphosphonate drug, Prolia (Denosumab), for osteoporosis. Introduction: Osteonecrosis of the jaw is a potential complication resulting from anti-osteoclastic medications. Osteonecrosis associated with the use of Prolia is reported to be infrequent. Management of these types of cases may be difficult. Methods: A patient reported a painful bony sequestrum occurring spontaneously on the mucosal surface on the lingual side

of the left mandible. A CBCT scan revealed spontaneous bone loss on the lingual side of tooth #18-20 and surrounding an implant on the site of tooth #30 in the mandible. The imaging showed breaks in the cortication along the lingual side of #18-20 and severe bone loss around the implant at site #30. The necrotic bony sequestrum was removed from the lingual mucosal surface of #18. On a follow-up visit, a small remaining bony sequestrum was treated. The implant was removed at site #30 followed by bone graft placement. Result: Following treatment, healing occurred at all sites. Conclusion: This case reports spontaneous osteonecrosis associated with use of Prolia aided by the use of CBCT imaging.

MULTIDISCIPLINARY

CC23 A Multidisciplinary Approach to Treating the Medically Compromised Patient. T. KULA III*, E. RAMOS, D. GEORGE, T. KULA JR. (Indiana University School of Dentistry)

As the patient population continues to live longer and has more complex medical conditions, consideration of the different disciplines in dentistry result in successful outcomes of care. A 72-year-old male presented to the IUSD dental clinics for an emergency examination of #11. He presented with unstable control of his blood pressure, atrial tachycardia, and a prosthetic knee replacement. Mobility and stamina limit his ability to tolerate long appointments. On examination, #11 was grossly carious with minimal coronal tooth structure remaining. The tooth was an abutment for a three unit fixed partial denture (FPD) from #11 to #13. Tooth #11 was asymptomatic. The radiograph demonstrated no interproximal bone loss and root length of approximately 21 mm. Tooth #12 was extracted approximately 45 years ago resulting in a large boney defect. The patient stated that he wanted to keep tooth #11 "at all costs." Treatment options were presented. Based on the patient desires and because of his current medical status, a multidisciplinary approach was employed to restore tooth #11. Treatment included interceptive endodontic therapy and forced eruption to expose adequate tooth structure to restore the tooth. A circumferential supracrestal fiberotomy procedure was performed weekly or biweekly to prevent alveolar bone from following the extruding tooth. The patient's dentition was restored with a FPD from #11 to #13. Total treatment time was approximately 6 months. The patient experienced no significant medical side effects from the treatment and was satisfied with the final result.

PERIODONTICS

CC24 Perils Of Piercing - A Case Report. K. RAJKUMAR*, E. RAMOS, S.B. BLANCHARD
(Department of Periodontics and Allied Dental Programs, Indiana University School of Dentistry)

Introduction: For more than two decades, oral piercing has been a common body piercing seen among young adults. Social pressures to appear "cool and fashionable" drive the desire for piercing. Intra-oral jewelry can cause adverse effects to both hard and soft tissues of the oral cavity including fractured teeth, prosthetic restorations and gingival recession. Numerous articles have reported gingival inflammation and recession but not periodontal attachment loss. In some cases, gingival recession is not present; therefore, a thorough periodontal evaluation is paramount. This case report will illustrate an approach to manage periodontal attachment loss associated with oral jewelry. Case Report: A 29 year-old Caucasian female presented to IUSD for a routine examination. Medical history was unremarkable. Patient reported a history of tongue piercing 8 years ago. She stopped wearing the oral jewelry 6 months prior to presenting to IUSD. Comprehensive oral and periodontal evaluations were completed. A probing depth of ~7mm accompanied by ~1 mm of gingival recession was noted on the lingual of tooth #24. The probing depths on rest of her dentition were less than 3 mm. No radiographic presentation of a bone defect in the region was evident. The treatment plan developed for the patient was guided tissue regeneration. On flap reflection, lingual bone dehiscence was noted on teeth #s 23, 24, 25. Calculus was found on the root

surface of #24. The roots were thoroughly planed and EDTA was applied. Enamel Matrix Derivative was applied to the root surface; some was also mixed with decalcified freeze-dried bone allograft (DFDBA) and grafted along the root surfaces of teeth #s 23, 24, and 25. The flap was repositioned and sutured. The post-operative period was uneventful. Patient was subsequently followed up. The probing depths at the end of 15 months were 2 mm circumferential on the mandibular anterior teeth. Conclusion: It is imperative to educate dental health care providers and patients concerning the potential adverse effects of oral piercing. A thorough dental and periodontal examination should be conducted on all patients with intra-oral piercings. Additionally, it is important to elicit a thorough history about any oral piercing in the past so as not to overlook any hidden periodontal defects. Early detection of periodontal damage secondary to intra-oral piercings is important so that corrective periodontal regeneration/ plastic surgery can be performed in a timely manner.

CC25 Effects of Platelet Rich Fibrin Matrix on Bone Regeneration and Soft Tissue Healing in Combination with Freeze Dried Bone Allograft in Ridge Augmentation: A Case Report. H. DUKKA*, M. GOSSWEILER, V. JOHN, S.B. BLANCHARD (Indiana University School of Dentistry)

Background: Platelet rich fibrin is regarded as a promising new biologic modifier in Periodontics. It is an autologous platelet and leucocyte enriched fibrin matrix containing multiple growth factors (VEGF, PDGF, EGF, IGF-1, bFGF, TGF- β 1, TGF- α , PAF), which play a vital role in attracting inflammatory cells, fibroblasts, as well as to stimulate collagen deposition, and endothelial budding, which leads to appropriate wound healing. Fibrin enhances soft tissue healing, rate and quality of bone regeneration and reduces postoperative hematoma formation. Aim: To describe a method of guided bone regeneration (GBR) using autogenous platelet rich fibrin matrix (PRFM) barrier with freeze dried bone allograft (FDBA) in conjunction with Ozone therapy. Description of the procedure: A healthy 61-year-old Caucasian female presented with non-restorable teeth #s 23, 24, 25 and 26 and wanted to replace them with dental implants. Upon clinical examination, the soft tissue around the teeth appeared inflamed and extremely erythematous. Radiographic examination revealed a large vertical defect requiring both horizontal and vertical ridge augmentation. The patient underwent the ozone therapy prior to extraction where ozonated water was injected into the soft tissues around lower anteriors to create a positive biochemical and physiologic change. Just prior to surgery, intravenous blood was collected in 10ml sterile tubes without anticoagulant and spun in a centrifuge at 3,000 revolutions for ten minutes. The teeth were then extracted atraumatically and FDBA was placed in the defect. The allograft was covered with three autogenous PRFM membranes. Primary closure of the surgical wound was achieved. Results: Postoperative healing was uneventful with exceptional soft tissue healing. A cone beam computed tomography at 2.5 months showed significant horizontal and vertical bone regeneration. Conclusion: GBR with FDBA and PRFM barrier membranes can be a viable option for accelerated soft and hard tissue healing. Primary closure is critical in achieving a successful outcome.

CC26 Sinus Augmentation and Simultaneous Dental Implant Placement by a Safer-Simplified-Sinus-Surgery-Technique. W. ZHANG*, S. TOWNS, S.B. BLANCHARD (Indiana University School of Dentistry, Graduate Periodontics)

Background: Pneumatization of maxillary sinus makes the placement of dental implants in posterior maxilla more challenging. Dental surgeons have predictably overcome these obstacles by performing sinus augmentation procedures. However, perforations of Schneiderian membrane are the most common complication of sinus augmentation procedures which may preclude implant placement. A Safer-Simplified-Sinus-Surgery-Technique (4ST) has been developed by Dr. Arthur Forrest for a less invasive lateral window sinus augmentation. This technique utilizes medical grade dense polyurethane sponges to

elevate sinus membrane. To date, there are no published reports concerning this technique for lateral window sinus augmentation with simultaneous dental implant placement. Aim: To describe a method of lateral window sinus augmentation with simultaneous dental implant placement with 4ST sponge technique. Case report: A 72 year old white female came to IUSD Graduate Periodontics for implant placement for missing #14. A pre-operative CBCT showed sinus pneumatization in the left maxillary sinus, and the height from the ridge crest to the floor of sinus was 5mm. The patient elected to have a lateral window sinus lift with simultaneous a dental implant placement under intravenous moderate sedation. The surgical sequences involved: A crestal incision was made from the mesial of #11 to the maxillary tuberosity area with a vertical releasing incision at the mesial line angle of #11. A full thickness flap was elevated buccally and a 6mm (W) X4mm (H) window in the lateral sinus wall was developed using a round low speed diamond bur. A piezosurgery handpiece with a diamond-coated tip was used to fully expose sinus membrane. Three dense polyurethane sponges were gradually introduced into sinus to elevate Schneiderian membrane. After elevation of sinus membrane, implant osteotomy was prepared with use of a surgical guide. Polyurethane sponges were removed and sinus cavity was grafted with Bio-Oss bovine bone mineral followed by placement of a Straumann 4.8 x 10 mm bone level implant. Additional Bio-Oss was placed around the implant, a resorbable collagen membrane (Collatape) was placed to cover the window, and flap was replaced and sutured using 4-0 Vicryl. Post-surgical radiographs showed good implant position with 10 mm of sinus floor elevation. Postoperative healing was uneventful with complete soft tissue closure at 2 weeks. The patient reported minimal post-surgical discomfort following the procedure. Conclusion: Lateral window sinus augmentation with simultaneous dental implant placement using medical grade polyurethane sponges (4ST) can be a simple, predictable treatment option with reduced complications associated with lateral window technique.