Payment for Orthodontic Treatment Parent-reported QOL of Teens

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Objectives: Medicaid funding for teens seeking orthodontic treatment is limited. This policy may have effects at a societal level and may lead to more severe and expensive dental and psychological sequelae. This project analyzes whether type of payment for orthodontic treatment in teens is associated with oral quality of life scores (TOQL) from the parents' and teens' perspectives. **Methods:** Participants were patients aged 10 to 18 years old enrolled in the Orthodontic clinic at BUGSDM and their parents who consented to complete TOQL questionnaires. Teens were classified into three groups based on their payment method: Medicaid, Medicaid and cash, and non-Medicaid. General linear modeling and regressions examined the association between esthetic scores and reimbursement type with TOQL scores reported by teens and their parents. **Results:** Data were from 127 teens and their parents, mean age 13, 55%male, 33.90%White, 25.42%Hispanic, 22.03%Black, 5.08 %Asian and 13.86% other races. The majority (55%) of the subjects either paid cash or used a private insurance; 32 % used Medicaid only and 14% Medicaid and cash. There was no association between esthetic scores and parent-reported TOQL and domain scores. Associations between payment type and parent-reported TOQL scores / domains are shown: PAYMENT METHOD Private / cash, Medicaid, Medicaid + cash P. TOQL – TOTAL SCORES were 8.48, 11.90, 15.07, p=0.06; EMOTIONAL DOMAIN: 5.74, 12.16, 13.12, p=0.06; ORAL HEALTH DOMAIN: 6.88, 8.90, 14.17,p=0.03; PHYSICAL DOMAIN: 3.83 8.11, 10.50, p=0.04; and SOCIAL DOMAIN: 20.41, 24.48, 33.18, p=0.21. **Conclusion:** Payment type is associated with only the oral health and physical domains of parent-reported quality of life of their teens. Supported by NIDCR Grant Nos. U54DE014264, U54DE019275, K24 DE018211 and Boston University Henry M. Goldman School of Dental Medicine.

Phosphorylation Sites Distribution by Mass Spectrometry for Bovine Dentin Phosphophoryn (BDP)

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Introduction: Dentin phosphophoryn (DPP), derived from the dentin sialophosphoprotein (DSPP) gene is the most abundant noncollagenous protein produced in dentin. DPP displays evidence of providing a role in the nucleation of hydroxyapatite formation within dentin during tooth development. Phosphoryn's highly repetitive acidic DSS sequence contains many phosphorylated serine residues that create a high binding affinity for Ca2+, which in the presence of Mg2+ may allow for the formation of hydroxyapatite (Cocking-Johnson D, Sauk JJ). The unerupted third molar present in the bovine oral cavity provides adequate protein concentrations of DPP during dentin formation for protein analysis. **Materials & Methods:** Utilizing techniques with mass spectrometry we will uncover the topographic distributions of phosphoserine residues within dentin phosphophoryn. Understanding these distributions of phosphorylation sites on DPP could engage further knowledge of the mechanistic actions provided by DPP in the formation of hydroxyapatite in dentin during tooth development. In addition, several mutations within the DSPP gene have exhibited pathological dentin diseases such as, dentinogenisis imperfecta types- II and –III and dentin dysplasia- II. (Macdougall M, et al. 2006) **Conclusions**: Further knowledge of dentin phosphophoryn could provide means of stabilizing these conditions within patients.

Cellular Changes of Stem Cells in 3D Culture

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Objectives: Mechanotransduction is particularly important in fields such as orthodontics, because the exact mechanism of translating the physical force into biochemical changes is unknown. Upon understanding cellular mechanosensing, future treatments could be established to promote faster and better Orthodontic treatment. Orthodontic treatment has long been recognized for its role in improving esthetics, function, and overall wellbeing. In recent years, however, orthodontic treatment has garnered additional importance in the delivery of oral care to an increasingly aging and/or health compromised patient population. Therefore, more efficient and controllable tooth movement is critical for progressing orthodontic treatment outcomes and providing optimal care. **Materials & Methods:** To determine the molecular effects and the associated cellular changes related with mechanical stress, wild type mouse bone marrow derived mesenchymal stem cells (MSCs) were used. MSCs are the precursor cells of osteoblasts and chondrocytes. The objective of the study was to evaluate cellular changes of MSCs in response to mechanical forces. These cells were subjected to either a control (no force) or force gel construct. Previously our research has determined there are in fact differences between cells when put under mechanical stress. Further confirming the 3D culture of MSCs is a useful model to study molecular mechanisms of mechanotransduction. However our prior research was only basic and preliminary; hence more thorough investigations were needed. In this current study we examined the proliferation of cells (Figure 5) and the apoptosis of cells (Figure 3) under mechanical stress by specific cellular markers (Ki67 and TUNEL). **Conclusions:** Understanding the cellular and molecular mechanisms regulating MSCs in response to mechanical stimulation will provide insights into designing novel strategies, such as locally injecting certain factors to facilitate orthodontic tooth movement.

Pathogenesis of Autoimmune Diseases and Dental Implications

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Objectives: The aim of this review is to summarize the current consensus on pathogenesis and the oral manifestations of systemic autoimmune diseases leading to a potential strategy to decrease the autoimmune diseases incidence by improving oral health. **Methods**: Pubmed using the terms autoimmunity, pathogenesis, Bechet's syndrome, rheumatoid arthritis (RA), systemic lupus erythematosus (SLE), Sjogren's syndrome, pemphigus and Oral lichen planus (OLP). **Results:** The presence of auto-reactive T and B-lymphocytes is physiological and termed autoimmunity. The transformation from autoimmunity to autoimmune disease is postulated to be triggered by environmental factors such as infections, chemicals, adjuvants and physical elements (UV radiation) in genetically susceptible persons. Naive T-cells develop into autoimmunity causing Th17 cells via the aid of multiple transcription factors, cells and cytokines including Treg, B-cells, Breg, IL-6, TGF-B, Foxp3, IL-23 and Th17. **Conclusion:** the pathogenesis and clinical presentations vary in each disorder, but the oral cavity presentation is often similar and many autoimmune diseases have oral ulcerations. It is essential for dental practitioners to be familiar with the pathophysiology and clinical presentations of these disorders and detect them early. Early detection will allow patients and healthcare providers to formulate a strategy to prevent or mitigate the disease progression and subsequent flare-ups.

Using a Cone Beam CT Scan to Evaluate Periodontal Bone Height

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Objectives: To compare the assessment of interproximal bone height using cone beam CT scans with that done using bitewing radiographs, in order to see if CT scans can be used instead of bitewings for the purpose of assessing bone height. **Methods:** CT scans were oriented and cropped to create the same image as a bitewing x-ray. The interproximal spaces with their CEJs to be measured were identified on both types of films. The distance was measured from each CEJ to the alveolar bone height directly below it. To account for the slope of bone and vertical bone loss, a straight line was drawn interproximally between the CEJs, and the height of the alveolus was measured to this line. Two interproximal spaces were measured on each radiograph/cone beam scan pair. Each radiograph was measured twice. **Results:** There was agreement within the two measurements of each bitewing/CT pair at 95% confidence, demonstrating that the methods are reliable. However, there are statistically significant differences between the two methods of measuring alveolar bone height. Differences between the two measurements ranged from 0.3 -0.6mm. **Conclusions**: Although the differences in measurements are statistically significant, they are not clinically significant. There is also inherent magnification in the bitewings which may lead to overestimation of distances. The detail of the CT scan can be affected by the resolution of the scan and the density of facial soft tissue. However, it is our conclusion that a CT scan is a sufficient instrument for measuring bone heights.

Impacts of Malocclusion are Different from Children's and Parent's Perspectives

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Objectives: Orthodontic treatment in children significantly improves their self-esthetic perception. The aim of this study is to measure social, functional, and esthetic effects of malocclusion by examining quality of life (QOL) scores from parents and children, and to determine how age, sex, and Index of Orthodontic Treatment Need (IOTN) scores affect QOL. Methods: Parents accompanying children (10-18 years old) receiving orthodontic care between 2010 to the present at Boston University School of Dental Medicine were asked to complete a questionnaire pertaining to their child regarding socio-demographics and QOL. ANOVA was used to determine the association between age, IOTN, and sex, with parent's and children's perception of child's QOL score. Results: A total of 141 parents accompanying their children completed the questionnaire. The children used in the study were on average 13.12 years old and were on average in 7 grade. Of the 123 subjects, 66 are male and 57 are female with 5.83% being Asian, 21.67% black, 26.67 Hispanic, 33.33% white, and 12.5% other. Quality of life scores were examined by age, sex, and IOTN score. Child-reported items in the social domain of QOL significantly differed by child sex and age (girls and older children with worse scores, $p=0.002$), parent-reported esthetic scores of children significantly differed by child age and IOTN score ($p=0.004$), and parent-reported oral problems in their children significantly differed by child age ($p=0.038$). Conclusion: Quality of life scores are affected differently by malocclusion in children based on child's age, sex, IOTN score, and parent vs. child report. To improve communications, it is important for practitioners to know which components of quality of life are salient to orthodontic patients and their parents. Supported by NIDCR Grant Nos. U54DE014264, U54DE019275, K24 DE018211 and Boston University Henry M. Goldman School of Dental Medicine.

Strength of Veneer Porcelain with Various Zirconia Thickness and Sintering

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Objective: Evaluate the effect of various firing cycles and zirconia thickness on the flexural strength of two veneering porcelains. **Methods:** Vita YZ zirconia blocks (Vita Zahnfabrik, Germany) were sectioned and sintered to provide slabs of 0.5, 1.0, and 5.0 mm thickness. Vita VM9 and e.max ceram porcelain (Ivoclar, Liechtenstein) were mixed with deionized water, condensed in a mold, and sintered: (1)According to the manufacturer's instructions; (2)Two cycles at peak temperature above the recommended value. A layer of carbon paint was applied to the zirconia to allow removal of the porcelain discs. Biaxial flexure strength was determined using an Instron, crosshead speed of 0.5mm/min. **Results:** Strength of e.max Ceram and VM9 at various cycles and zirconia thickness N=10 Peak Temp -Zirconia thickness Flexural Strength MPa Significant Difference 800°C - 0.5 Zr 120.4 ± 15.5 A 800°C – 5.0 Zr 114.6 ± 24.9 AB 800°C – 1.0 Zr 114.4 ± 14.5 AB 780°C - 1.0 Zr 107.6 ± 18.7 ABC 760°C - 1.0 Zr 100.8 ± 10.6 BCD 780°C – 0.5 Zr 99.0 ± 15.6 CD 780°C - 5.0 Zr 98.1 ± 15.6 CD 760°C – 0.5 Zr 94.4 ± 19.8 CD 760°C - 5.0 Zr 88.5 ± 12.3 D VM9: Peak Temp - Zirconia thickness Flexural StrengthMPa Significant Difference 940°C – 1.0 Zr 122.6 ± 13.9 A 940°C – 0.5 Zr 121.2 ± 14.6 A 960°C – 5.0 Zr 120.4 ± 15.4 A 940°C - 5.0 Zr 118.7 ± 12.1 A 920°C - 1.0 Zr 116.2 ± 12.7 A 960°C – 5.0 Zr 114.6 ± 14.5 A 960°C -1.0 Zr 114.4 ± 14.5 A 920°C – 0.5 Zr 110.3 ± 15.4 A 920°C - 5.0 Zr 107.9 ± 21.7 A ANOVA and Tukey analysis, p=0.05. **Conclusion:** Firing cycle and zirconia thickness have a significant affect on the strength of e.max ceram.

Unmet Preventive Dental Care Needs Among Children with Special Health Care Needs

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Objectives: To evaluate the status of unmet preventive dental care needs among Children with Special Health Care Needs (CSHCN) in the US. **Methods:** We analyzed data of 40,242 CSHCN from the National Survey of Children with Special Health Care Needs (NS-CSHCN) 2009-2010 conducted by the Centers for Disease Control and Prevention. Children with unmet preventive dental care needs were identified as those with perceived the needs but without having received such care. Other variables included were: Age, race, language spoken at home, insurance status, federal poverty level, parents' education, area of living, and the number of special care needs conditions. Bivariate and multivariate weighted analysis was conducted to evaluate the access to preventive dental care among CSHCN. **Results:** Overall, 10% of CSHCN had unmet preventive dental care needs. Children of older age (15-17 y/o), African-Americans, non-English speakers, uninsured, from low socioeconomic level, living with low educated parents were significantly associated with having unmet preventive dental care needs. Furthermore, the percentage of children with unmet preventive dental needs increases gradually as they have more special care needs conditions. In the logistic regression model, insurance status, federal poverty level and the number of special care needs conditions remain significant determinants for having unmet preventive dental care among CSHCN. **Conclusion:** Our findings indicate there are significant disparities in the access to preventive dental care among CSHCN. Programs and policies targeting the barriers to preventive dental care to eliminate the disparities and improve the oral health of CSHCN need to be explored and implemented.

N-glycosylation Signaling Pathways in Oral Squamous Cell Carcinoma

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Oral Squamous cell carcinoma (OSCC) or oral cancer accounts for majority of head and neck cancers and ranks as the sixth most common cancer in the world. OSCC belongs to the most understudied cancers and little is known about molecular mechanisms underlying its etiology and progression to metastasis. Likewise, no molecular biomarkers have been identified to inform decisions about either timely diagnosis or treatment. A hallmark of cancer is the enhanced posttranslational modification of cell surface proteins with complex N-glycans. Our studies have shown that induced protein N-glycosylation via activation of the core N-glycosylation-regulating gene, DPAGT1, is associated with reduced E-cadherin adhesion, as well as deregulation of several oncogenic signaling pathways, including Wnt/β-catenin and Hippo. Modest increases in DPAGT1 expression are associated with dramatic amplification of Wnt/β-catenin activity and increased expression and nuclear localization of the Hippo effectors TAZ and YAP. Increased Wnt/β-catenin and TAZ/YAP nuclear activity promote cell proliferation, stem cell-like properties and drug-resistance. **Objectives:** The goal of this study was to align the expression and localization of DPAGT1 , surface presentation of complex N-glycans, β-catenin and TAZ/YAP with development and progression of oral cancer *in vivo* from dysplasia to OSCC. **Methods:** Human oral tissues from different stages of OSCC pathogenesis were characterized for DPAGT1/β-catenin/YAP/TAZ expression and localization and correlated with cell surface expression of complex N-glycans by PHA lectin staining and with expression of primitive of cell surface markers, CD44, CD24 and CD29. **Results** showed that high DPAGT1 expression and nuclear TAZ became increasingly associated with disorganized E-cadherin junctions as oral epithelium progressed from mild to severe dysplasia to OSCC. This correlated with increasing expression of cell surface complex N-glycans and CD44. **Conclusion:** These studies indicate that DPAGT1/β-catenin/TAZ and high PHA staining represent novel signatures for OSCC development.

Skeletal and Dentoalveolar Effects of Class II Treatment Modalities in Patients Treated with Suresmile: A Comparative Retrospective Pilot Study

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Objectives: To determine and compare the skeletal and dentoalveolar effects of multiple class II treatment modalities in Class II Division I malocclusions. **Methods:** A retrospective study was conducted using lateral cephalograms from Conebeam CT's taken on patients treated with Suresmile. Subjects that were included in this study exhibited a skeletal discrepancy of > or equal to 4 degrees > or equal to 4mm of overjet, bilateral end-on molar occlusion or greater, and ranged in age from 8-14 years old. Subjects were divided into groups based on class II treatment modality and included distalization using TADs(n=12), Class II elastics (n=6), FORSUS appliance (n=4), and a combination group including distalization using TADs + Class II elastics and FORSUS + Class II elastics (n=6). Pre-treatment and post-treatment lateral cephalograms were traced and comparative outcomes of the parameters measured from the data were analyzed using a one-way analysis of variance along with multiple comparison of each method to show all possible comparisons between each group. **Results:** We found that none of the groups showed statistically significant results in parameters that significantly contribute to comparative differences in correction of Class II Division I malocclusions except between the multiple comparisons of U1-NA in degrees between the FORSUS and Combination group. A P value of .05 was considered statistically significant. **Conclusions:** There is a significant difference between the change in upper incisor proclination between The FORSUS and combination group. Though the overall results of this study showed no statistical significance for the parameters measured, trends from means in IMPA and L1-NB in degrees and millimeters for TADS, Class II elastics, and FORSUS show positive contribution towards Class II Division I correction dentally, but at the cost of compensation. Mean ANB decreased in all groups and U1-NA (mm) decreased in TADS and FORSUS groups. This indicates the need for further investigation to confirm significance and comparative conclusions.

Role of Ellis-van Creveld Syndrome2 (Evc2) in Craniofacial Development

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Background: Ellis-van Creveld (EvC) syndrome is a rare chondro-ectodermal dysplasia with an autosomal recessive trait affecting bone and cartilage growth. EvC patients have mutations in either EVC or EVC2 gene, both of which are located on chromosome 4 in a head-to-head configuration. **Objective:** There were several cases reported that abnormal craniofacial bone phenotype was observed in EvC patients; however, it is currently unknown whether mutation of EVC or EVC2 gene causes such craniofacial bone phenotypes. Our objective is to investigate and characterize the craniofacial phenotype using Evc2 knockout (KO) mice. Methods: Evc2 KO mice were used in this study as an animal model of EvC syndrome and craniofacial bone development/phenotype in these mice was investigated in comparison to controls, i.e. wild type and heterozygous mice. Lateral cephalometric radiographs and analysis were conducted on three postnatal groups [one (n=15), three (n=11), and six (n=9) weeks old]. The expression pattern of Evc2 was investigated; in addition, onset and levels of both proliferation and apoptosis in chondrocytes were identified in the developing embryos [E15.5-E18.5]. **Results:** Our data showed that the postnatal bone growth deficiency in KO mice was found in areas where expression of Evc2 was observed. Growth rate of craniofacial bones in KO mice was reduced to 72-79 % of that of controls at the tested time points. Notably, growth of certain bones including nasal bone, palatal bone and premaxilla was more affected in KO than in the controls. Furthermore, there was a remarkable change in facial bones' spatial relationship to the cranial base and vault. We also found an earlier onset of apoptosis and proliferation defects in chondrocytes in KO compared to controls. **Conclusions:** Evc2 is required for craniofacial bone development and deficiency in Evc2 leads to specific facial bone growth defect due to imbalance of cellular proliferation and cell death.

Comparison of Nasal Airway Dimensions in Patients with Vertical and Horizontal Growth Patterns: A Cone Beam Computed Tomography Evaluation

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Background: Obstructive sleep apnea (OSA) is one of the most commonly studied disorders involving the airway and can have serious consequences on overall health and quality of life. The role of the pharyngeal airway in OSA has been extensively studied. However, the role the nasal airway plays is less well understood. The importance of the airway on facial development and development of malocclusion has been well established. Although greater visualization of anatomy and better diagnosis of disorders are now possible, classifying the extent of airway disorders cannot be done without a reference point. It is therefore important to establish normative values of the airway size and dimensions from which comparisons can be made. Although recent studies have begun to establish normative values of the pharyngeal airway, normative values of the nasal airway have yet to be established. The establishment of normative values in patients with different skeletal growth patterns will allow for comparison between these groups and aid we may be able to determine if patients that present with certain skeletal growth patterns have a higher risk of developing sleep apnea or other breathing-related disorders. **Aim:** To establish normative values of the nasal airway using Cone-Beam Computed Tomography (CBCT) and compare these values in patient with different skeletal growth patterns. **Material and Methods:** The sample included 106 patients ages 9-50 who had previous CBCT scans taken as part of routine orthodontic treatment. Scans were analyzed using Anatomage Invivo5 software to identify 3-dimensional cephalometric landmarks. Patients were classified by their anteroposterior growth patterns as Class I, Class II, and Class III based on the Steiner analysis. Patients were further classified by their vertical growth patterns as hyperdivergent or hypodivergent based on both the mandibular plane angle and the Jarabak anaylsis. The maximum height (mm) and maximum width (mm) of the nasal cavity were measured through visual inspection. **Results:** Final statistics are still pending, but will be presented on the poster. However, there is a general trend that shows that patients with a hyperdivergent, or more vertical, growth pattern have smaller dimensions of the nasal cavity than patients with a hypodivergent growth pattern. The trend also shows that the nasal airway tends to enlarge with growth. Increases in height and width were noted during the pubertal growth spurt and continued to increase throughout the third decade. **Conclusions:** Patients presenting for orthodontic treatment that display vertical growth tendencies may have smaller dimensions of the nasal cavity. These patients may require further evaluation and referral to specialists before orthodontic treatment can initiated. Additionally, the orthodontic treatment plan may be altered to include growth modification in attempt to control a vertical growth pattern. The nasal cavity may play more of a role in OSA than previously thought and further research into this area is needed.

The Validity of Radiation Free 3D Imaging as an Orthodontic Diagnostic Tool

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Background: Cephalometrics has been the foundation of orthodontic diagnosis for many years. However, some orthodontists maintain that for many routine orthodontic cases a lateral cephalogram is not necessary. Aim: The purpose of this study is to develop a novel method of orthodontic diagnosis and treatment planning that utilizes three dimensional images of the face and teeth in lieu of traditional orthodontic records and in particular, the lateral cephalogram. **Material and Methods**: 9 orthodontic patients were treatment planned by 10 different orthodontists using a set of standard orthodontic records by filling out a diagnosis and treatment planning questionnaire. The same patients were retreatment planned by the orthodontists after 2 weeks using 3D records. Internal consistency of treatment planning decisions (extraction versus non-extraction and orthognathic surgery versus non-orthognathic surgery) were examined by Kappa statistics and Kendall's tau statistics as applicable. **Results:** A total of 89 patient treatment plans were analyzed at two different time points using standard and 3D records. Following evaluation of standard records, 33 treatment plans (37.1%) were designated as needing extractions and 11 (12.4%) were deemed to require orthognathic surgery. Following evaluation of 3D records, 20 treatment plans (22.5%) were designated as needing extractions and 4 (4.5%) were deemed to require orthognathic surgery. Kappa statistic for extraction versus non-extraction was 0.45 ($p<0.0001$) showing poor reliability. Kappa statistic for orthognathic surgery versus non-surgery was 0.22 ($p=0.02$) indicating poor reliability. **Conclusion:** Preliminary data indicates poor reliability between the standard and 3D records. Intra-examiner treatment planning reliability needs to be assessed to determine whether differences are due to the records or due to orthodontist inconsistency. One explanation as to why fewer extractions and surgical cases were treatment planned is that the 3D records allowed the orthodontists to better assess the profile needs of the patient. If the differences are due to the records, we believe that the 3D records could still be used as a valuable adjunct to standard orthodontic records. Further testing is needed to arrive at a valid conclusion.

Directed Differentiation of Induced Pluripotent Stem Cells Into Endothelial-like Cells

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Efficient derivation of endothelial cells (ECs) from embryonic stem cells (ESCs) or induced pluripotent stem cells (iPSCs) likely will require a stepwise approach, termed “directed differentiation” involving the replication of the key commitment stages found during embryonic development. **Methods:** We developed a directed differentiation protocol for producing ECs from iPSCs. In stage 1, hiPSCs were cultured in a media supplemented with Activin, BMP4, bFGF to induce primitive streak (PS) differentiation. In stage 2, BMP4 is required for differentiation of the cells to mesodermal cells expressing KDR (receptor for human vascular endothelial growth factor, VEGF). In stage 3, addition of VEGF and bFGF generated EC progenitorlike cells with 10-15% efficiency and these cells could be sorted to purity based on coexpression of KDR, CD31 (PECAM) and CD144 (VECadherin) by day 12. **Results:** The sorted cells maintained proliferative capacity and could be expanded in endothelial maintenance media. Compared to control ECs, our purified iPSC derived ECs, similarly continued to express CD31, CD144, KDR, and von Willebrand factor by qPCR and flow cytometry, and were functionally capable of taking up AcLDL and forming tubes in an in vitro angiogenesis assay. Finally our protocol was then applied to generate disease specific ECs from iPSCs derived by reprogramming skin fibroblast of a patient with pulmonary hypertension due to BMPR2 mutations. **Conclusion:** Our results demonstrate a novel platform for the de novo generation of patient specific cells with phenotypic characteristics similar to ECs.

Comparison of Esthetic Brackets Made of Composite, Porcelain and Alumina

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Objective: Determine difference of properties between composite (MZ100), porcelain (MARK II) and alumina brackets (Fascination ® 2) under different conditions. **Methods:** Transcend Brackets were mounted on an orthodontic wire and scanned for reproduction via CAD/CAM using a Cerec inLab machine. Test specimens were made of Vita MARK II and 3M/ESPE MZ100 blocks. Brackets were tested using torque and tipping modes via custom fixtures and an Instron universal testing machine. Torque was exerted on the brackets via 0.014 x 0.014 inch stainless steel arch wire. Testing was done both wet and dry. Bracket materials were tested for resistance to staining using coffee and turmeric. Specimens were immersed in test solutions and stored in an incubator at 37◦C for 1 month, then washed and dried and checked for color change using a spectrophotometer. The effect of ultrasonic and brushing with toothpaste was also evaluated. **Results:** The failure load means ± SD (gm-mm) and failure angle means ± SD (degrees) are: Material MZ100 Mark II Alumina Condition Dry Wet Dry Wet Dry Wet Torque angle 77.0±15.0 60.0±6.5 56.0±5.7 47.0±13.0 79.0±6.9 37.21±12.9 Torque load 2504±494 2525±591 1406±170 1006±237 1900±267 2142±557 Tip angle 69.0±22.0 55.0±13.0 114.5±0 Tip load 8990±1954 2333±1227 11905±2220 **Conclusions:** Although MZ100 brackets and MARK II brackets were significantly weaker than the alumina brackets in their resistance to either second order (tipping) or third order (torsional) activations, they proved to be strong enough to withstand the suggested magnitude of clinical archwire torsional and tipping forces under dry and wet conditions. The results of staining test showed that the MZ100 specimens were visibly discolored with turmeric.

Delayed Peripheral Blood Neutrophil Apoptosis in Diabetes and Chronic Periodontal Disease

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Apoptosis plays a critical role in the host immune response and resolution of inflammation. Several studies have shown delay of spontaneous neutrophil apoptosis in diabetes and chronic periodontitis but it remains unknown whether both condition combined will affect neutrophil apoptosis. **Objective:** To compare the neutrophil apoptosis between patients with diabetes and chronic periodontitis to healthy individuals. **Methods:** The study included 73 patients consisted of type II diabetes (DM) (n= 16), chronic periodontitis (CP) (n= 15), combined diabetes with chronic periodontitis (DM+CP) (n= 21) and healthy volunteers (H) (n= 21). Heparinized venous blood was obtained. Neutrophils were isolated by centrifugation in Histopaque 1119, and Histopaque 1077 (Sigma). The neutrophil-rich layer was collected and contaminating erythrocytes were lysed with a hypotonic NH4Cl buffer (Sigma). Cells were incubated and maintained in RPMI-1640 supplemented with bovine fetal serum by 2 to 24 h. Neutrophil apoptosis levels were determined by flow cytometry with Tunel assay (BDbioscience). Caspase-3 activity was measured by colorimetric assay (Invitrogen). Statistical analysis was performed using ANOVA with LSD post-hoc test. **Results:** Spontaneous Neutrophil apoptosis reached 50% in 8.8h in healthy subjects and 10.9h, 13.0h and 14.2h in diabetes, chronic periodontitis and combination respectively. In 12h neutrophil undergoes apoptosis 66.5% in healthy group and 52.5%, 49.1% and 48.7% in DM, CP and DM+CP respectively. Significant difference was observed between healthy group compared to all the diseased groups ($p<0.05$). In addition, the caspase-3 activity assay, healthy group shows the highest result in every time points. In 12h, the level of caspase-3 activity in healthy subjects was significantly higher compared to all patients with diabetes. **Conclusion:** Our results suggested that spontaneous neutrophil apoptosis is decreased and delayed in diabetes and chronic periodontitis, both condition united further impaired the spontaneous apoptosis which promote tissue injury and compromise the resolution of inflammation. Supported by NIH Grant NIH2340.

Lysyl Oxidase Like-2 Mediates CCN2-Stimulated Gingival Fibroblast

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Background: CCN2/CTGF is elevated in fibrotic gingival overgrowth (GO) lesions; and stimulates collagen accumulation in human gingival fibroblasts in vitro. CCN2/CTGF up-regulates lysyl oxidase like-2 (LOXL2). Objective: Investigate LOXL2 as a mediator of CCN2/CTGF enhanced collagen accumulation & cell proliferation. **Methods:** Primary human gingival fibroblasts were cultured from three different healthy patients. Up-regulation of LOXL2 by CCN2/CTGF protein was confirmed by Western blot. Cell proliferation assays (Cyquant) in response to 24-hour treatment with 200 ng/ml rhCCN2/CTGF or vehicle in LOXL2 shRNA-, non-target-, and empty virus transduced gingival fibroblasts cells were performed. Growth curves (7 days) were conducted. Effects of pharmacologic inhibitors of LOXL2 enzyme activity on collagen accumulation (Sirius Red assay) and cell proliferation assays (Cyquant and growth curves) were carried out plus/minus CCN2/CTGF in human gingival fibroblasts from three different patients. Finally, effects of exogenously added active human LOXL2 on collagen accumulation & cell proliferation was determined. **Results:** CCN2/CTGF increased lysyl oxidase like-2 (LOXL2) protein in human gingival fibroblasts by 3.5-fold after 6 hours. LOXL2 shRNA lentivirus reduced the LOXL2 mRNA and protein by 80 – 95%. Knockdown of LOXL2 strongly inhibited both basal and CCN2/CTGF-stimulated collagen accumulation ($p < 0.05$). Proliferation assays demonstrated a marked 50% decrease in CyQuant and growth curve assays in LOXL2 shRNA knockdown cells. Pharmacologic inhibition of LOXL2 enzyme activity reduced basal- and CCN2/CTGF-stimulated cell proliferation (40% and 50%) and collagen accumulation (49%). Recombinant active LOXL2 increased collagen accumulation by ~30% ($p<0.05$) **Conclusion:** LOXL2 is critically required for both gingival fibroblast proliferation and for collagen accumulation in the presence or absence of CCN2/CTGF. CCN2/CTGF up-regulation of LOXL2 (mediated by JNK Map kinase) is a major mechanism of CCN2/CTGF activity relevant to gingival overgrowth etiology. The mechanism by which LOXL2 stimulates gingival fibroblast proliferation is under investigation. Supported by NIDCR DE 011004.

Isolation and Purification of a Gluten-Degrading Enzyme from Human Gastrointestinal Tract-derived Bacteria

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Introduction: Celiac disease is a T-cell mediated-inflammatory disorder of the small intestine caused by the intake of gluten-containing food. Gluten proteins are difficult to digest by mammalian digestive enzymes. Surviving immunogenic gluten domains cause inflammatory immune responses in predisposed individuals. We have previously presented evidence that gluten can be degraded by a fecal microbial strain (FA-10) at low pH. Enzymes with activity at low pH are of interest since they may be able to digest gluten during gastric transport. **Aims:** 1) To isolate and purify the enzyme from strain FA-10 culture supernatant by DEAE and MonoQ chromatography; 2) To determine cleavage activities towards mixed gliadins and a gliadin-derived immunogenic 33-mer peptide. **Results:** Proteins were separated by DEAE chromatography into 11 fractions of which fractions 2-6 showed activity in a gliadin zymogram. Those fractions were pooled and proteins separated by MonoQ chromatography into 8 fractions. One of those fractions showed a clear band in the gliadin zymogram gel and a single major band in an SDS gel. The enzyme preparation degraded 250 µg/ml of the gliadin-derived 33-mer peptide by 83.3%, 79.1%, 67.7% and 41.7% respectively after 0.5 h, 2 h, 5 h and 24 h incubation at 37▪C. **Conclusion:** The enzyme derived from strain FA-10 was isolated and semi-purified by sequential anion-exchange chromatography. The discovery of a low pH-active gluten-degrading enzyme provides new therapeutic perspectives for the treatment of celiac disease. These studies were supported by NIH/ NIAID grants AI087803 and AI101067

Xerostomia and Salivary Hypofunction in Vulnerable Elders: Complications and Management

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Xerostomia and Salivary Hypofunction in Vulnerable Elders: Complications and Management Michael R. Dion, Bing Liu, Laura Kaufman, Steven L. Karpas, and Judith Jones Background: Xerostomia is the subjective sensation of oral dryness while salivary gland hypofunction (SGH) is the objective observation of reduced salivary flow. It has been a common practice to use the term dry mouth to describe both conditions. Dry mouth is a major complaint in vulnerable elderly patients (VE) with compromised general health and limited resources. **Objective:** The purpose of this article is two-fold: first, to review existing studies describing oral complications of xerostomia and SGH; second, to review clinical management options in VE. **Methods:** The authors searched PubMed (May 1989-November 2012) for English-language articles related to the complications and management of dry mouth and determined whether the studies identified met the inclusion criteria of this review. Data were summarized from the included publications and presented herein. Over 400 articles were identified initially; articles included in the references were based on the consensus of all authors. Only articles targeting the relief of dry mouth, either xerostomia or SGH, as the primary goal were included. **Results:** Common complications reported from xerostomia and SGH include caries, oral fungal infection and compromised quality of life. Epidemiological data points to possible correlation between dry mouth and other oral conditions, including periodontal disease, soft tissue lesions, and changes in oral functions in VE. The current management of both xerostomia and SGH remains palliative. Preparations of oral hygiene products have been improved for the routine application for VE. Saliva substitutes with natural salivary components such as mucins and enzymes appear to be well tolerated by VE; the clinical outcomes have been favorable. There is heightened interest in remedies with minimal side effects, such as acupuncture and electro-stimulation. Limitations and future research directions have been discussed. **Conclusions:** Dry mouth adversely impacts the general health of vulnerable elders, including oral structures, oral functions, and quality of life. Due to its complex etiology and the health concerns of VE, management of dry mouth in VE is challenging. Current clinical regimens may provide limited relief of symptoms. Large-scale, long-term and well-designed clinic trials are needed to assess the efficacy of treatment in VE. Clinical Implications: Oral status of VE should be evaluated periodically for signs and symptoms of salivary dysfunction. Effective management of dry mouth in VE is crucial to general well-being and needs to be modified on an individual basis, which can best be achieved by the team work of physicians, dentists, caregivers and patients. Key Words Vulnerable elder, xerostomia, salivary hypofunction, dry mouth, saliva substitute. Supported by Geriatric Fellowship

The Correlative Role of Epiprofin and *Tbx1* in Tooth Proliferation and Differentiation

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Introduction: In tooth morphogenesis, the dental epithelium and mesenchyme interact reciprocally for growth and differentiation. This interaction is critical in the formation of the proper number and shapes of teeth. Epiprofin (*Epfn*) and *Tbx1* have been previously identified as transcription factors involved in the process of tooth development. **Materials & Methods:** To identify the role each transcription factor has in tooth development, *Epfn* and *Tbx1* wild-type, heterozygous, and knockout mice were studied. Further analyses regarding the role of both transcription factors utilized western blots, qPCR, and immunofluorescence. These analytical procedures were conducted to quantify the levels of both transcription factors throughout the 18 day embryonic developmental period. **Results:** *Epfn -/-* mice developed an excess number of teeth, enamel deficiencies, dentin defects. *Tbx1 -/-* mice exhibited a reduced number of teeth as well as enamel defects. The altered signaling events may underlie critical functions carried out by *Epfn* and *Tbx1* affecting the differentiation, proliferation and maturation of ameloblasts and odontoblasts during tooth development. We aimed to study *Epfn* and *Tbx1* in the incisors of developing mice in order to better elucidate their functionality as well as identify any possible interactions between the two transcription factors. **Conclusion:** In the future, we hope to apply our findings regarding *Epfn* and *Tbx1* into both further understanding certain diseases including DiGeorge Syndrome and Ectodermal Dysplasia, as well as potentially elucidate particular points of interest for future targeted therapy. Supported by the National Institute of Dental and Craniofacial Research Summer Dental Student Award Program and the Boston University Henry M. Goldman School of Dental Medicine Office of Pre-doctoral Research

The Standardization of a Non-radiation Technique for Orthodontic Diagnosis

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Objective: Orthodontic diagnosis currently involves examining the patient and taking photographs, radiographs, and study models to assess positions of the jaws and teeth. Those measurements are then compared to norms which were selected by orthodontists over 50 years ago. Historically, orthodontists have treated patients to conform to those norms regardless of the final soft tissue outcome. The aim of this study is to standardize the use of three dimensional photographs along with a panoramic radiograph to diagnose orthodontic problems. Method: Approximately 200 female models were screened from modeling agencies. The 20 most attractive models were chosen by 40 lay people using a visual analog scale. Intraoral scanning was done using a 3M Lava scanner(3M,St. Paul, MN). 3D facial scans using a Canfield 3D facial scanner(Canfield Imaging Systems, Fairfield, NJ) were taken in natural head position, both at smiling and at rest. The two scans were then combined. Inclusion criteria for the models: 1. Class I molar and canine occlusion: 2. Crowding or spacing less than 2mm. 3. OB and OJ between 2-3mm. 4. No missing teeth except for 3rd molars 5. No crossbite 6. No openbite 7. Age between 18 and 35years 8. Considered attractive by laypeople. Inclusion criteria for the lay people: 1- No medical or dental training. 2-Age at least 18 years. 3-Acceptable vision and understanding to answer the survey. **Results:** Dentoskeletal and soft tissue norms are created using the three dimensional facial and intraoral scans to aid the orthodontists in diagnosis and treatment planning. Preliminary results with the sample of 11 models show variations in facial proportions. Conclusion: By standardizing a technique of combining 3D extraoral photographs with 3D intraoral scans, orthodontist are able to diagnose many orthodontic cases without the need for additional records.

The Effect of Serum on in Vivo Early Microbial Colonization of Tooth Enamel

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The oral environment surrounding the oral soft and hard tissues is determined by the biochemistry of saliva. Therefore, all molecular events occurring on exposed oral surfaces are influenced by the composition of this oral fluid. A particular exception applies to the space immediately above the gingival sulcus facing the tooth surface. This is due to the presence of gingival fluid, a well known entity in oral biology characterized by the slow outflow of a serum-like fluid. Once emanated from the gingival sulcus, this fluid mixes with saliva, creating a serum-saliva gradient. The gradient originates at the orifice of the gingival sulcus and becomes completely salivary in nature at some distance from the sulcus. It is well established that the biofilm of tooth surfaces is the major culprit for caries and periodontal disease development. The critical early event of biofilm formation is the specific interaction of microorganisms with the acquired enamel pellicle, a thin protein film coating tooth enamel. This pellicle structure dictates the specific adsorption of biofilm microorganisms. We hypothesize that the microbial binding specificity may change depending on the type of adsorbed proteins present being derived either from serum or saliva. **Objective:** To investigate the effect of serum proteins binding to enamel surfaces on the adsorption of the early microbial colonizers**. Methods:** 5 mL of blood were obtained by venipuncture and its serum fraction prepared. Pellicle and dental biofilm were removed from the buccal surfaces of teeth comprising first molar to first molar in both arches. Two- thirds of the cleaned coronal surfaces of incisor, canines and pre-molars of the upper and lower right arch were coated with 5 µL of serum derived form the same subject. As a negative control, the incisors, canines and pre-molars of the upper and lower left arch were coated with 5 µL of water. The teeth were exposed to the normal oral environment for a specified period with the subject refraining from the intake of food or drinks, except water. After exposures to oral conditions for 0, 2, 4 and 6 hours biofilm material was harvested and its microbial composition analyzed by the Human Oral Microbial Identification Microarray. **Preliminary Results:** Adequate coverage of the enamel surfaces was achieved with 5 µL of serum. Microbial DNA was successfully prepared from the collected sample of a single subject. The intended enrollment for this study will be 10 subjects. Microbial composition data of serum and water covered tooth surfaces will be subjected to the paired Wilcoxon Signed-Rank Test and Spearman Correlation Coefficient analysis. Significance will be set at $p > 0.05$. **Conclusion:** This will be the first study to investigate whether serum proteins in the in vivo formed acquired enamel pellicle will lead to biofilm characteristics conducive to periodontal disease development. The data could also provide new information on prevention and therapeutic management of the disease. Supported in part by NIH grants: AI087803, DE05672, DE07652 NSERC grant: 371813 CIHR grants: 106657, 97577 CFI-LOF grant: 25116 WLS is recipient of a CIHR New Investigator Award grant # 113166.

Cellular Uptake of rLOX-PP Involves Phosphatidylinositol 3-kinase (PI(3)K) Dependent, HSPG Mediated Macropinocytosis, Caveola-dependent Endocytosis.rLOX-PP alkalizes endosomes via modulating endosomal pH

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rLOX-PP is an excellent candidate to cross cellular membranes and to treat cancer. The lysyl oxidase propeptide (LOX-PP) is derived from pro-lysyl oxidase (Pro-LOX) by biosynthetic proteolysis. It was identified as an inhibitor of the transformed phenotype in NIH 3T3 fibroblasts overexpressing c-H-ras; and in breast, lung and pancreatic cancer cells with mutant RAS genes. rLOX-PP attenuates fibronectin-stimulated integrin signaling and migration in breast cancer cells. In addition, rLOX-PP inhibits FGF-2-induced DNA synthesis and cell growth in developing normal primary and MC3T3-E1 osteoblasts and DU145 prostate cancer cells by inhibiting FGF-2 binding to FGFR1. Although, several intracellular targets and molecular mechanisms of rLOX-PP have been identified, rLOX-PP uptake pathways have not been previously investigated. Here we demonstrate for the first time that the major uptake pathway for rLOX-PP appears to be PI(3)K and HSPG-dependent macropinocytosis in PWR-1E, DU145, PC3, SCC9, MDA-MB-231 cell lines. A secondary pathway appears to be dynamin and caveola dependent uptake employed by PC3, SCC9, MDA-MB-231 cell lines. rLOX-PP uptake through the plasma membrane occurred in an energy-independent manner. ATP-depletion increased rLOX-PP translocation through plasma membrane at 37 °C by lowering endosomal pH (pHi), the cationic property of rLOX-PP provides buffering capacity to rLOX-PP at both high and low pHs. We suggest its buffering capacity facilitates rLOX-PP endosomal escape into the cytoplasm enabling its observed interactions with cytoplasmic targets and ultimately its nuclear uptake. Supported by NIH grant R01DE014066.

Morphometric Analysis of Facial Structures and Its Relationship To Attractiveness

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Background: Orthodontists, as well as plastic and oral and maxillofacial surgeons have a special interest in facial beauty and consider improvement of facial esthetics an important goal. Since the general public is in fact the one who receives these improvements, orthodontists must study facial beauty, as is perceived by the public. Traditionally, orthodontic treatment planning originated at the teeth and the skeletal features of the face and subsequently, the effect of treatment on soft tissues were evaluated. Contemporary orthodontics, on the other hand, evaluates the soft tissue first in an 'outward-in' fashion to determine what skeletal and dental treatment is needed to bring about the desired soft tissue changes. In order to accurately assess the craniofacial morphology a three-dimensional analysis is required and Geometric Morphometrics is the method of choice in assessment of the craniofacial shape. **Methods:** We presented 108 photographs (obtained during routine protocol for orthodontic diagnosis and treatment planning) of skeletally mature Caucasian subjects to 30 college students. The judges rated the attractiveness, symmetry and distinctiveness of these faces on a 1-7 Likert Scale and estimated the subjects' ages. The same images were previously rated by 169 orthodontists based on their 'facial balance'. A pixel-based pattern-recognition model was then used to evaluate these faces. To discover the factors contributing to judgments of attractiveness, we used Partial Least Square (PLS) analysis. We used the same model to evaluate an additional 215 photographs from the same database to expand our subject pool and increase the odds of encountering "attractive" faces from which three-dimensional normative values would be derived. We digitized 98 hard tissue landmarks and semilandmarks on CBCT scans of these subjects. Selected points were assessed in the sagittal, coronal and axial slices to confirm accurate landmark localization in all three planes of space. Then, CBCT scans of the subjects were analyzed for their morphometry using Geomorph. The same program was used to obtain an average shape from the attractive group on which Procrustes superimposition of random CBCT scans can be done. **Results:** We found that the pixel-based pattern recognition model's attractiveness ratings were congruent with both the orthodontists' and laypeople's ratings. Therefore, using this model was helpful in expanding our subject pool of attractive faces from which a normative database was derived. A multivariate regression analysis between the shape data and attractiveness scores of the most attractive and the least attractive groups showed a significant relationship ($R^2=0.084$). We also found that on average groups occupy different areas in shape space ($p=0.001$). In other words, the most attractive and the least attractive groups are very different in shape. **Conclusion:** Geometric Morphometrics proved to be a significant tool in analyzing skeletal shapes and establishing a baseline that can be used in diagnosis and planning of orthodontic and orthognathic surgical treatments. Considering that most 2D cephalometric analyses are based on norms derived from patients with 'normal occlusion', our 3D analysis that is based on facial attractiveness supports the contemporary 'outward-in' trend in orthodontic and orthognathic surgery fields.

Assessement of Potential Gluten-like Properties of Salivary Proline-rich Proteins

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Background: Celiac disease is a T-cell mediated-inflammatory enteropathy caused by dietary gluten. Structurally gluten are unusual with respect to their high content in proline (P) and glutamine (Q) residues, features that are related to their toxicity. Interestingly the high P/Q content is also found in a group of proteins present in human saliva, the proline-rich proteins (PRPs). PRPs are synthesized and secreted by salivary glands into oral cavity and reach the gastrointestinal tract through swallowing. We hypothesize, based on the similar structures of dietary gluten and salivary PRPs and their shared destination of the gastro-intestinal tract, that salivary PRPs may exhibit gluten (specifically, gliadin)-like properties. **Aim:** To study if parotid saliva (PS), which contain PRPs, could induce T-cell inflammatory responses in peripheral blood mononuclear cells (PBMCs) preparations from celiac disease patients and to investigate if parotid salivary proteins can interfere with the gliadin-triggered immune response through competition. **Methods:** Parotid saliva was collected from refractory celiac disease (RCD), celiac disease (CD), and healthy control (HC) subjects. PBMCs were freshly isolated from blood of CD patients. Gliadins and PS proteins were pre-digested with pepsin, trypsin, and modified with transglutaminase-2 to mimic in vivo deamidation. Gliadin or PS proteins (each 100 µg/ml) were added to PBMCs (2X106 cells/ml) and incubated for 24 h at 37°C. Cytokine levels (TNF-α, IL-10) were tested in the cell supernatant. To study if parotid saliva proteins could interfere with gliadin-triggered immune responses, 25 µg/ml, 50 µg/ml and 100 µg/ml PS were mixed with 100 µg/ml gliadin, and cytokine release was determined after 24 h incubation. **Results:** Parotid saliva samples were collected from 4 HC, 4 CD, and 6 RCD patients. PBMCs were isolated from two CD patients and cytotoxic cytokine levels were successfully induced with the digested and deamidated gliadin sample. Preliminary data showed that when gliadins were mixed with PS proteins, the cytokines levels were reduced by PS protein in a dose-dependent manner. **Conclusion:** This is an ongoing study where the potential augmenting or dampening effects of salivary PRPs on the cytotoxicity of gliadins is being investigated. The results of this investigation may reveal salivary PRP domains that could affect gliadin toxicity and may play a role in the establishment of tolerance to gluten and gliadins early in life.
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