

ORAL ABSTRACT PRESENTATIONS

#1- 5/1/2017 - 8:00 AM

IN VIVO ANTINUCLEAR ANTIBODY POSITIVITY OF ORAL EPITHELIUM MAY INDICATE SYSTEMIC CONNECTIVE TISSUE DISEASE.

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While anti-nuclear autoantibody (ANA) granular staining of epithelial cells is characteristic of chronic ulcerative stomatitis (CUS), it is currently unknown whether ANA staining of oral biopsies is also indicative of other autoimmune diseases. Moreover, the ANA staining patterns present in oral epithelium and the correlative H&E findings are poorly understood. We hypothesized that ANA positivity in oral epithelium may be an adjunctive diagnostic marker of systemic connective tissue diseases. We performed a retrospective study to describe the pattern of in vivo ANA in oral epithelium, to correlate this with H&E findings, and determine whether patients with these findings had a co-existing systemic connective tissue disease. We examined 72 consecutive cases from our lab collected between 2013 and 2016 that showed ANA staining on direct immunofluorescence (DIF) of oral biopsies. Submitting clinicians were contacted to determine what follow-up was performed and the final diagnosis that resulted. Follow up clinical information was available for only 10 of the cases. These patients had diagnoses of autoimmune diseases including rheumatoid arthritis, systemic lupus erythematosus, systemic sclerosis, and CUS. Immunoglobulin G was the most common immunoreactant (71/72 cases) while speckled nuclear staining was the most common in vivo ANA pattern (56/72). Thirty-four cases had an H&E biopsies and most were non-specific chronic mucositis (28/34) or lichen planus (23/34). Based on our findings, we recommend that patients who show in vivo ANA staining of oral mucosal biopsies should be further investigated for presence of any systemic autoimmune connective tissue disease.

#2- 5/1/2017 - 8:12 AM

A PILOT STUDY OF PD-1 AND PD-L1 EXPRESSION IN A SPECTRUM OF ORAL DYSPLASIAS AND ORAL SQUAMOUS CELL CARCINOMAS

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As our understanding of head and neck cancer increases, personalized therapy with targeted approaches is becoming more common. Recently, the FDA approved the checkpoint blocking antibody pembrolizumab to treat advanced head and neck squamous cell carcinoma (SCC). This cancer immunotherapeutic monoclonal

antibody blocks the interaction of the programmed cell death - 1 (PD-1) receptor on T-cells from interacting with the programmed cell death receptor ligand - 1 (PD-L1) produced by tumor cells. Normal cells utilize the PD-1/PD-L1 pathway to control immune recognition. Tumor cells exploit this pathway by upregulating their expression of PD-L1, which results in downregulation of the immune response and ultimately apoptosis of inactive T-cells. To better understand PD-L1 and PD-1 interactions within oral tissue, we used immunohistochemistry on 12 severe oral epithelial dysplasias from high-risk sites and 10 oral SCCs, two of which were recurrent. The cases were additionally characterized based on inflammatory phenotype. Results showed 7 cases had epithelium that stained positive for PD-L1. Of the 7 positive cases, 5 were SCCs and 2 were dysplasias with a heavy inflammatory infiltrate including germinal center formation. Seventeen cases had PD-1 positive immune cells. Interestingly, 15 cases had immune cells that stained positive for PD-L1. Although the majority of severe dysplasia cases did not express PD-L1 on epithelial cells, elucidating the staining characteristics of immune cells may prove to be beneficial in better understanding these lesions and potential treatments.

#3- 5/1/2017 - 8:24 AM

A RETROSPECTIVE 20-YEAR ANALYSIS OF PROLIFERATIVE VERRUCOUS LEUKOPLAKIA AND ITS PROGRESSION TO MALIGNANCY AND ASSOCIATION WITH P16INK4A IMMUNOHISTOCHEMICAL STAINING

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Proliferative verrucous leukoplakia (PVL) is a multifocal form of leukoplakia that commonly affects elderly females with higher propensity for malignant transformation. In this study, we evaluated the malignant potential of PVL in the UF Oral Medicine Clinic patients seen over the last 20 years and evaluated its association with p16INK4a expression. An IRB approved retrospective search was performed in the UF College of Dentistry Oral Medicine Clinic Database from 1994 to 2016 for all patients with a clinical and/or biopsy proven diagnosis of PVL. Exclusion criteria included cases with one or more excisional biopsy with either no follow-up or a follow-up of less than 3 years. Demographics, history of tobacco use, medical conditions, and histological diagnoses were recorded for each patient. Analysis of the selected 20 cases showed a wide age range of 34-87 years, with a mean age of 62.75 years. A female predominance with a female-to-male ratio of 2.3:1 was noted. Mean follow-up was 6.6 years, with an average of 4.4 biopsies per patient during this period. Ten PVL patients had additional systemic

conditions, 6 of whom developed malignancy. Multiple sites of involvement were reported for most patients, gingiva being the most common location.

Approximately 50% of PVL cases progressed to squamous cell or verrucous carcinoma. Immunohistochemical staining for p16INK4A was largely negative, with a 50-65% positivity observed in 3 cases which progressed to malignancy, whereas 7 cases showed only a basal cell layer staining pattern. Therefore, no definite association between PVL and p16INK4a expression was noted. Due to the high malignant transformation potential of PVL, early recognition, close clinical follow-up, and multiple biopsies are essential for proper management.

#4- 5/1/2017 - 8:36 AM

ALVEOLAR SOFT PART SARCOMA METASTATIC TO THE MANDIBLE: A REPORT AND REVIEW OF LITERATURE

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Objective: Alveolar soft part sarcoma (ASPS) is a rare neoplasm which accounts for less than 1% of all soft tissue sarcomas. Most cases of ASPS occur in adolescents and young adults, with women more often affected than men. ASPS usually presents as an indolent, painless mass, with a predilection for soft tissue of the lower extremities. Here, we report the case of a 29-year-old woman with ASPS metastatic to the mandible. **Clinical Presentation:** The patient had a known 12-year history of ASPS, which first presented as a left psoas mass at the age of 17. She subsequently developed metastases to her left kidney, right lung, right shoulder, lumbar spine, and liver over her disease course. The patient was referred to an oral surgeon with a chief complaint of increasing pain in her right mandible of approximately six months duration. Imaging studies revealed a mixed radiolucent-radiopaque lesion in the body of the right mandible near teeth #28-30. **Intervention and Outcome:** Fine needle aspiration (FNA) of the mass showed malignant cells consistent with alveolar soft part sarcoma. Following this diagnosis, the patient underwent a tumor debulking procedure for palliation. **Conclusion:** Overall, there are 54 documented cases of ASPS (including this case) occurring in the oral cavity, of which only four represent cases of metastasis. Of these, only two are metastatic to the mandible. To the best of our knowledge, this represents only the 5th documented case of ASPS metastatic to the oral cavity, and more specifically, the 3rd documented case of mandibular metastasis. Awareness of such a clinical presentation is important for the clinician, especially in younger female patients or patients with relevant medical history who present with lytic jaw lesions of unknown etiology.

#5- 5/1/2017 - 8:48 AM

SOFT TISSUE ANGIOLEIOMYOMA (VASCULAR LEIOMYOMA): A REPORT OF 29 NEW CASES AND REVIEW OF THE LITERATURE

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Oral angioleiomyoma (OAL) are uncommon tumors of the oral mucosa. The purpose of this study was to report the clinicopathologic features of 29 new cases of OAL and to review the English literature. **Materials and Methods:** Cases diagnosed as angioleiomyoma and vascular leiomyoma were identified from the files of Harvard School of Dental Medicine from 2005 to 2015. **Results:** There were 17 males and 12 females and the median age was 54.0 (range 20-76). The most common location was the lip (22 cases, 75.9%) followed by the palatal mucosa (5 cases, 17.2%) with two cases on the buccal mucosa (6.8%). Pain was reported in only one case and all tumors were less than 2 cm in greatest dimension. All tumors were composed of a circumscribed but nonencapsulated mass of smooth muscle cells in a slightly whorled pattern. The tumor cells had abundant pale, eosinophilic and vacuolated cytoplasm, and spindle and fusiform vesicular nuclei with inconspicuous nucleoli. Dilated vascular spaces were present lined by a single layer of endothelium. There were 95 cases identified in the literature comprising 63 males, 32 females. The median age was 46.0 (range 10-76). The lip was affected in 47.8% of cases followed by the palatal mucosa (18.9%), the buccal mucosa (8.2%) and the tongue (7.5%). **Conclusion:** OAL is an uncommon benign smooth muscle tumor most commonly seen in the lip in adults, more often males, in the 5th and 6th decade.

#6- 5/1/2017 - 9:00 AM

CLINICOPATHOLOGIC FEATURES OF SCURVY IN AN AUTISTIC CHILD WITH A VITAMIN C DEFICIENT DIET

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Objective: Vitamin C or ascorbate is required for collagen synthesis and iron absorption. Vitamin C also has antioxidant properties. Ascorbate acts as a cofactor in the activation of lysyl and prolyl hydroxylases from inactive precursors. Subsequently, the enzymes facilitate procollagen folding. Insufficient dietary ascorbate leads to qualitative and quantitative type IV collagen abnormalities resulting in capillary fragility and osteopenia, thus producing the characteristic clinical phenotype of scurvy. These features include gingivitis, musculoskeletal dysfunction and perifollicular hemorrhages. Iron absorption is indirectly affected,

leading to anemia. Individuals with disordered eating habits and malnutrition, such as the elderly, alcoholics and autistic children, are at risk for developing scurvy. Clinical Presentation: An 11 year old autistic child with a limited diet presented for evaluation of hemorrhagic gingiva, musculoskeletal weakness and purpuric skin lesions. The clinical presentation suggested scurvy. Intervention and Outcome: Laboratory evaluation and gingival biopsy were performed, confirming the diagnosis of scurvy. Supplementation with a multivitamin and vitamin C was provided. Biopsy revealed ulcerated mucosa, extravasated red blood cells and edematous granulation tissue, consistent with scorbutic gingivitis. Iron deficiency anemia, serum vitamin C level of 0 mg/dl and deficiencies in vitamins A and D were identified. Vitamin supplementation revealed clinical and symptomatic improvement at ten days and three weeks. Conclusion: Scurvy should be considered in the differential diagnosis of hemorrhagic, hyperplastic, necrotic gingivitis, anemia and musculoskeletal and dermatologic abnormalities, in the clinical setting of autism and dietary deficiencies.

#7- 5/1/2017 - 9:12 AM

SPINDLE CELL AMELOBLASTIC CARCINOMA WITH EVIDENCE OF EPITHELIAL-MESENCHYMAL TRANSITION IN TUMOR PROGRESSION

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Spindle cell ameloblastic carcinoma is a rare histological variant of ameloblastic carcinoma, with only 9 cases reported in the literature. This malignancy demonstrates histologic features of ameloblastic differentiation, accompanied by significant cytologic atypia and a prominent spindle cell population. Our purpose is to report two additional cases of this rare entity in young people, and to present histological and immunohistochemical evidence of epithelial-mesenchymal transition in tumor cells in these two cases. Our first patient, a 24-year-old male, presented with an expansile lesion in the right maxilla accompanied by pain and loose teeth. The second patient, a 12-year-old female, presented with a 1-year history of an expansile radiolucent lesion in the left posterior mandible.

Histological examination of both cases revealed malignant epithelial neoplasms showing ameloblastic differentiation, with spindle cell populations that appeared to arise from the epithelial component. Immunohistochemical studies for cytokeratins and vimentin demonstrated a transition from expression of cytokeratin without vimentin staining in tumor cells of epithelial morphology, to expression of vimentin and loss of cytokeratin staining in the spindle cell component. Sox-2 showed focal positivity in both components, and Ki-67 stained approximately 6-

10% of the tumor cells. Both patients underwent surgical resections of their tumors with unilateral neck dissections in May of 2015. The first patient also underwent adjuvant radiation therapy. Currently, 21 months after their surgeries, neither patient has shown evidence of recurrent disease.

#8- 5/1/2017 - 9:24 AM

ASSESSMENT OF BIOLOGICALLY AGGRESSIVE, RECURRENT GLANDULAR ODONTOGENIC CYSTS FOR MASTERMIND-LIKE 2 REARRANGEMENTS: HISTOPATHOLOGIC AND FLUORESCENT IN SITU HYBRIDIZATION FINDINGS OF ELEVEN CASES

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Objective: Glandular odontogenic cyst (GOC) is a rare developmental cyst with propensity for aggressive behavior and recurrences. While distinct, some histopathologic features of GOC may overlap with those of intraosseous mucoepidermoid carcinoma (IMEC), an uncommon malignant primary salivary gland neoplasm of the gnathic bones. Previous studies have shown that GOCs lack the specific mastermind-like 2 (MAML2) gene rearrangements found in IMECs. This study is designed to evaluate the presence of MAML2 rearrangements in biologically aggressive GOCs and their possible potential for transition to IMEC. In addition, this study reviews histopathologic guidelines for the diagnosis of GOCs. Methods and Results: Eleven cases of recurrent GOCs were evaluated utilizing break apart fluorescent in situ hybridization (FISH) analysis for the presence of MAML2 gene rearrangements. Nine cases were from the mandible and one from the maxilla. The site of one case was not identified. The M:F ratio was 5:6 and the mean age at diagnosis was 55.27 years (36-72 years). Six patients presented with initial expansion of the jaw. Only one of the eleven cases showed a positive MAML2 fusion transcript. Conclusion: Our findings demonstrate lack of molecular evidence to support transition of aggressive, recurrent GOCs to IMEC. We also conclude that the established minimum of six histopathologic parameters is still the best indicator for the diagnosis of GOC. Furthermore, odontogenic cysts exhibiting mucous elements and MAML2 rearrangements while lacking the classic GOC histopathologic parameters should not be regarded as GOC.

#9- 5/1/2017 - 9:36 AM

WNT SIGNALING IN ORAL CANCER INITIATING CELLS

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Introduction: Oral squamous cell carcinoma (OSCC) is one of the leading cancers in the world. Patients with distant metastases of OSCC have 5-year survival rates of 38%. Cancer initiating cells (CICs) are proposed to explain the high recurrence rate of OSCC. CICs are specialized tumor cells that can self propagate, form heterogeneous clones and escape conventional therapy. Lineage tracing can be employed to identify individual clones of CICs and their behavior. Methods and results: To characterize CICs in premalignant lesions and OSCC, we traced the CICs based on their Wnt/ β -catenin activity. 4-NQO carcinogen was used to generate premalignant lesions and OSCC in Axin2-CreER;YFP mice. Tamoxifen was then applied to induce Cre activity, which lead to labeling of CICs. Histopathologic evaluation revealed malignant progression similar to human OSCC, i.e. hyperkeratosis, epithelial dysplasia and OSCC. Immunohistochemical studies for β -catenin revealed loss of membranous staining in basal cells in epithelial dysplasia. In OSCC, increased nuclear expression of β -catenin was noted, especially at the invasive front. In addition, immunofluorescence demonstrated co-expression of β -catenin and LEF1 in OSCC, suggesting activation of Wnt/ β -catenin signaling. Increased Axin2 fluorescence reporter was visualized in basal cells in OSCC. Lineage tracing was performed in 3-D organoid culture. Axin2 positive cells were capable of forming organoids, demonstrating stemness in the Wnt-responsive CICs. Conclusions: Wnt-responsive CICs in OSCC contribute to its malignant progression. Further research is needed to clarify the role of Wnt-responsive CICs in recurrence and therapy resistance.

#10- 5/1/2017 - 9:48 AM

STAT6 RELIABLY DISTINGUISHES SOLITARY FIBROUS TUMORS FROM MYOFIBROMAS

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Introduction: Solitary fibrous tumors (SFT) and myofibromas (MF) historically have been considered along the same morphologic spectrum as each other under the nonspecific umbrella term, hemangiopericytoma along with other pericytic/myoid tumors. While current evidence shows clear distinction between the two entities, they frequently remain in the histopathologic differential diagnosis. This is especially true for smaller incisional biopsies, as in cases from the oral cavity. STAT6 immunohistochemistry recently has been established as a reliable method to detect solitary fibrous tumor; however, limited knowledge about reactivity to STAT6 in MFs is available in the literature. Methods: After IRB approval, all SFTs and MFs were collected from the University of Florida Oral and Maxillofacial Pathology Biopsy Service between the years 1994 and 2016. The

original hematoxylin and eosin slides, as well as any accompanying immunohistochemistry slides, were reviewed. Immunohistochemistry for STAT6 was performed on all 35 tissue samples, and the findings were analyzed. Results: 10 Cases of SFT and 25 cases of MF were identified from the archives. Most cases were from the oral cavity. All SFTs expressed STAT6 nuclear reactivity (9 diffuse and strong, 1 patchy), while no cases of MF showed nuclear expression of STAT6. Weak, non-specific reactivity in one MF case was found. Conclusion: STAT6 is a dependable marker that can be used to differentiate SFTs from MFs.

#11- 5/1/2017 - 10:00 AM

CARTILAGENOUS METAPLASIA IN ODONTOGENIC KERATOCYST (OKC): CASE SERIES AND IMMUNOHISTOCHEMICAL ANALYSIS

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Cartilaginous metaplasia in the wall of an OKC is an established yet rare occurrence. It is important to recognize this feature to avoid misdiagnosis as a chondroid tumor. We describe the largest series of cases with the objective to quantify the clinicopathologic features and to elucidate the nature of the cartilage. Fourteen lesions in 13 patients (1 recurrence) of OKCs exhibiting cartilaginous metaplasia in the wall were identified between 1997 and 2016.

Immunohistochemical (IHC) analysis was performed on two cases. Eight of 13 cases occurred in the maxilla with 4 occurring anteriorly and 2 involving the maxillary sinus. Five cases occurred in the mandible, 3 anteriorly, including the recurrence. The average age was 55 with a range of 16 to 86 years old. Ten of 13 cases occurred in males. All 3 female cases occurred in the anterior maxilla. The chondroid areas were frequently identified in zones of inflammation near the OKC lining. The proportion of cartilage to cyst was variable. IHC for p63, GFAP, EMA, CK5/6, and actin were performed on two cases to elucidate the nature of the chondroid material. CK5/6, EMA, and p63 positivity was localized to the epithelial lining. GFAP and actin were negative in the cartilaginous areas. Our findings suggest that compared to conventional OKCs, there is a slight increase in age and predilection for the anterior jaws when cartilaginous metaplasia is observed. GFAP negativity suggests that the chondroid material may not represent actual cartilage. Cartilaginous metaplasia in the walls of OKCs should not be mistaken for a cartilaginous neoplasm.

#12- 5/1/2017 - 10:12 AM

A COMPARATIVE CYTOMORPHOMETRIC STUDY OF BUCCAL MUCOSAL SMEARS OF CIGARETTE SMOKERS AND NASWAR (NICOTIANA TABACUM) USERS

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ABSTRACT Objective: To determine and compare cytomorphometric changes in buccal mucosal cells of cigarette smokers, naswar users and non-users/smokers. **Materials and methods:** Cellular diameter CD, nuclear diameter ND and nuclear to cytoplasmic ratio N/C ratio were assessed in buccal smears taken with wooden spatula from clinically normal mucosa of smokers, naswar users and control group. The sample size was 99 subjects of ages 15 yrs-60yrs, divided into three groups (33 each group) as M, S and N i.e control, smokers and naswar users respectively. Slides were stained with three stains Hematoxylin and Eosin Stain, Giemsa Stain and Papanicolaou Stain. **Results:** The cytomorphometric variables were measured by using stage and ocular micrometers. The mean cellular diameter of group M, S and N was 43.8 μ m, 54.3 μ m and 42.7 μ m respectively. The mean nuclear diameter of M, S and N was 9.97 μ m, 12.6 μ m and 11.8 μ m respectively. And the mean N/C ratio of group M, S and N was 1:4.4, 1:4.3 and 1:3.5 respectively. The mean differences between CD, ND and N/C ratio in all three stains, among the three groups, S, N and M was found to be statistically highly significant i.e $p = 0.001$ on ONE WAY ANOVA. While, on Post hoc tukey test between S and N, CD and N/C ratio were highly significant $p=0.000$ while ND was not significant. While between S and M, CD and ND both were significant and showed $p=0.000$ while N/C ratio was not significant. In comparison between N and M, ND and N/C ratio were found to be significant while CD was not significant. **Conclusion:** The cytomorphometric changes assessed by this study depicts only cause effect relationship with smoking and naswar use. Association of these changes with dysplasia or pre-malignancy needs further verification with the help of specific immune-markers.

#13- 5/1/2017 - 10:24 AM

LOSS OF CALPROTECTIN (S100A8/A9) ASSOCIATES WITH POOR EPITHELIAL DIFFERENTIATION AND INCREASED EGFR EXPRESSION IN HEAD AND NECK SQUAMOUS CELL CARCINOMA (HNSCC).

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Background: Calprotectin, a heterodimeric complex of the calcium-binding proteins S100A8 and S100A9 (S100A8/A9) encoded on chromosome 1q21,

activates the G2/M cell cycle checkpoint, inhibits carcinoma cell migration and invasion, and suppresses tumorigenesis in vitro and in vivo. Calprotectin mRNA and protein expression decreases in poorly differentiated HNSCC and is associated with lower survival rates. Signaling through EGFR is also considered a negative prognosticator for HNSCC. Aims: 1) To investigate changes in S100A8/A9 expression during the continuum of oral carcinogenesis and 2) address whether EGFR expression is affected by S100A8/A9 status. Materials and methods: FFPE sections of 34 HNSCCs including 16 well-differentiated (WD), 8 moderately-differentiated (MD), 4 poorly-differentiated (PD) and 6 basaloid HNSCCs, and 15 premalignant epithelial dysplasias were immunohistochemically stained for S100A8 and S100A9. EGFR immunoexpression was studied in 17 HNSCCs. Results: Strong cytoplasmic and nuclear S100A8/A9 staining was seen throughout normal oral mucosal epithelium (NE) except for the basal cell layer which was consistently S100A8/A9 negative. Dysplastic lesions showed significantly less calprotectin expression than NE. WD HNSCCs strongly expressed calprotectin, while S100A8/A9 was progressively lost in MD, PD and basaloid tumors. EGFR expression was limited in S100A8/A9-high HNSCCs. Conversely, S100A8/A9-low carcinomas appeared to upregulate EGFR. Conclusions: In HNSCC, S100A8/A9 expression positively associates with the level of squamous differentiation of malignant cells and inversely correlates with EGFR expression. S100A8/A9-associated down-regulation of EGFR could explain better survival of patients with S100A8/A9-high tumors.

#14- 5/1/2017 - 10:36 AM

COMPOSITION OF IMMUNE CELL AGGREGATES AT THE INVASION FRONTS OF ORAL CANCER ASSOCIATED WITH CLINICAL OUTCOMES
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OBJECTIVES: Evading immune destruction from tumor microenvironment has been proposed as one of the hallmarks of cancer. However, little is known in oral cancer (OC). The objective is to investigate the immune cellular composition of invasion fronts (IFs) of OC and associated outcome. **METHODS:** As an exploratory study, two 5- μ m serial sections from the cross-section of 12 highly annotated OC samples were used for 2 sets of multi-color immunostaining with antibodies against: CD8, CD163, FoxP3, CD25; and PD-L1, PD-1, CD3, CD20 (Deeley Research Lab, Victoria, BC). **RESULTS:** PD-L1 tumor-cell expression was heterogeneous in 9 OCs, especially at the IFs. PD-L1- tumors had a better outcome with no nodal disease. In addition, PD-L1+ immune cells with dendritic

morphology were seen in all cases. In areas with interconnecting clusters of PD-L1+ immune cells we observed different proportions of CD25+ immune cells and CD8+-cells co-localizing within T-cell clusters. We did not observe a relationship between the number of CD8+-cells and the outcome; however, clusters of PD-1+ immune cells were commonly seen in patients with poor outcomes. Aggregates containing both expanded B-(CD20) and T-(CD3) cells at the IFs seemed to have a better outcome. CD163 was not exclusively expressed in macrophages. Various patterns of CD25 expression were also found at the IFs of the tumor cells of 11 OCs, including membranous, cytoplasmic, and dot-like. The latter was more evident in poorly differentiated tumors, while well-differentiated tumor nests expressed little to none CD25. CONCLUSION: Immune cellular composition at TME, either pro-inflammatory or immune suppressive, are associated with clinical consequences. Further investigation is warranted to shed light of the underlying biology.

#15- 5/1/2017 - 10:48 AM

DOWNREGULATION OF DMBT1 PROMOTES INVASION IN HEAD AND NECK CANCER.

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Deleted in malignant brain tumors 1 (DMBT1) is a tumor suppressor that is downregulated in multiple cancers. Objective: In the present study, we investigated the expression, function and mechanism of regulation of DMBT1 in human head and neck cancer. Methods: Using laser capture microdissection we isolated epithelium from head and neck cancer and normal tissue and used quantitative RT-PCR to quantify DMBT1 transcripts. The regulation of DMBT1 was investigated by genetic and biochemical approaches in human head and neck cancer cell lines. The function of DMBT1 was investigated using in vitro and in vivo approaches. Results: DMBT1 is downregulated in head and neck cancer compared to normal epithelium. DMBT1 expression is inversely correlated with EZH2, an oncogene that promotes invasion in head and neck cancer. Furthermore, EZH2 silences DMBT1 via histone and promoter methylation. In head and neck cancer cell lines with stable downregulation of EZH2, suppression of DMBT1 rescues the invasive phenotype. Conclusions: DMBT1 is a tumor suppressor that inhibits invasion and is silenced in HNC. (This work was supported by NIDCR grants DE022567 and DE019513)

#16- 5/1/2017 - 11:00 AM

GENE EXPRESSION PROFILE OF AMELOBLASTOMA

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Odontogenic tumors are a heterogeneous group of lesions that arise from the tissues derived from the tooth forming apparatus that display a range of microscopic patterns and clinical behavior. The most common is the ameloblastoma, a benign, locally infiltrative odontogenic tumor that on rare occasions may undergo malignant transformation. Surprisingly little is known about the molecular biology of most odontogenic tumors but the best known aberration is the presence of a BRAF V600E mutation in 40-60% of mandibular ameloblastomas. The goal of this project is to identify the gene expression profile of ameloblastoma and other common odontogenic neoplasms using laser microdissection and RNA sequencing, with the goal of finding novel markers for ameloblastoma and other odontogenic tumors. We analyzed gene-expression patterns of 18 odontogenic tumors. The epithelial component and the stromal component are obtained using laser capture microdissection, resulting in a total of 42 samples. Smart-3SEQ, a newly designed RNA sequencing method, is then used to prepare the cDNA libraries. 42 libraries pool are then sequenced using SMART-3SEQ. A total of 196.6 million reads are obtained, with an average of 4.68 million reads per sample. Genes with less than 20 reads are excluded from our study. Significant clusterings of several candidate genes are seen in ameloblastoma. We then performed differential expression analysis of the RNA-seq data using DEseq2. Based on the adjusted p-value (<0.05), >500 statistically significant candidate genes are found. We have identified a list of candidate genes that are preferentially expressed in ameloblastoma. These results may provide a framework to identify useful markers for diagnosis of odontogenic tumors.

#17- 5/1/2017 - 11:12 AM

A CLINICOPATHOLOGIC AND SURVIVAL ANALYSIS OF HEAD AND NECK SOFT TISSUE SARCOMAS (HNSTS) WITH KNOWN GENETIC ABNORMALITIES

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Background. HNSTS are rare and diagnostically challenging without the advent of ancillary testing to exclude more common diagnoses such as melanoma and carcinoma. An increasing number of STS are now classified based on their defining molecular genetic abnormalities and thus the criteria for diagnosing HNSTS have evolved from morphologic and immunohistochemical tools alone.

Method. We carried out a clinicopathologic and survival analysis focusing mainly

on HNSTS with known genetic abnormalities, treated and followed-up at our institution. Results. There were 147 patients (81 M and 66 F, mean of 21-yrs), 48% being of pediatric age (<18 yrs). The neck soft tissue (27%) was the most common site, followed by paranasal sinuses (16%). The mean tumor size was 4.6 cm. The tumor types included 53 embryonal RMS, 34 SS18-fusion positive synovial sarcoma, 33 FOXO1-positive alveolar RMS, 8 EWSR1-positive Ewing sarcomas (ES), 8 spindle-sclerosing RMS (SRMS) [3 MYOD1 mutated], 7 MPNST (all except 1 with loss of H3K27me3) and 4 CIC-rearranged round cell sarcomas. The 2, 5 and 10-year OS rates were 91%, 77% and 64%. Diagnosis (MPNST, followed by SRMS), large tumor size, stage and recurrence were adverse predictors on OS ($p<0.0001$). ES had the best outcome. There was no significant effect on OS with age and gender. Conclusion. HNSTS represents a spectrum of many tumor types, each characterized by specific genetic alterations. The application of molecular diagnosis is critical not only for a refined classification but also providing information on prognosis and guiding therapy. Our results showed that patients with MPNST had the worst outcome overall, while SRMS were the most aggressive among RMS, proving the utility of molecular investigation as an effective diagnostic adjunct.

#18- 5/1/2017 - 11:24 AM

COMPROMISED SOCKET HEALING IN RATS UNDER ZOLEDRONIC ACID TREATMENT AFTER EXTRACTION OF TEETH WITH PERIODONTITIS: INSIGHTS INTO THE PATHOGENESIS OF OSTEONECROSIS OF THE JAWS

A. Soundia, D. Hadaya, N. Efsandi, I. Gkouveris, R. Christensen, S.M. Dry, O. Bezouglaia, F.Q. Pirih, N. Nikitakis, T. Aghaloo, S. Tetradis, Department of Pathology and Laboratory Medicine, David Geffen School of Medicine at UCLA, Los Angeles, CA, USA, Division of Constitutive and Regenerative Sciences, UCLA School of Dentistry, Los Angeles, CA, USA, Dept of Oral Pathology and Medicine, School of Dentistry, National and Kapodistrian U. of Athens, Greece. Objective: Osteonecrosis of the jaws (ONJ) is a rare but severe complication of antiresorptive medications, such as bisphosphonates. To gain insight into the role of tooth extraction and dental disease in ONJ pathogenesis, this study investigated socket healing after extraction of healthy or periodontally compromised teeth in rats treated with zoledronic acid (ZA). Methods: WistarHan rats were divided into Groups 1 and 2 (extraction of healthy or periodontitis-involved teeth in vehicle-treated animals) and Groups 3 and 4 (extraction of healthy or periodontitis-involved teeth in ZA-treated animals), respectively. Animals were pretreated with vehicle or ZA for a week and periodontitis was induced. Four weeks later, the second molar teeth were extracted and sockets were allowed to heal for another

four weeks. Results: Radiographically, extraction sockets in Groups 1, 2 and 3 demonstrated normal healing; contrary, incomplete socket healing was noted after periodontal teeth extraction in ZA-treated rats of Group 4. Histologically, persistent inflammation and extensive osteonecrosis were seen in Group 4; picosirius red staining revealed distinct disorganization of the collagen network with lack of collagen fiber insertion in the necrotic bone. Immunohistochemistry revealed increased collagen type III and decreased collagen type I expression in Group 4; cells positive for MMP9, MMP13 and \pm -SMA expression were present at the areas of epithelial invagination and adjacent to osteonecrotic bone. Human biopsies from ONJ patients showed similar immunohistochemical results. Conclusion: Our data emphasize the importance of periodontal disease and tooth extraction in ONJ pathogenesis and help delineate an altered profile in wound healing markers during ONJ development.

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MAINTENANCE OF HEAD AND NECK CANCER STEM CELL-LIKE CELLS BY CD44/SMURF1 SIGNALING

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Head and neck squamous cell carcinoma (HNSCC) is among the most invasive malignancies, and the 5-year survival rate stands at approximately 50%. Recent studies in HNSCC suggest that a subpopulation of tumor-initiating cancer stem cells (CSC) is responsible for metastatic invasion and drug resistance. CD44, a receptor for hyaluronic acid (HA), is a known CSC marker in HNSCC, but its role in maintaining CSC populations is not known. We previously reported that SMURF1 inhibition of BMP signaling is essential for maintaining a CD44-high CSC-like population in HNSCC. Here, we investigated the mechanisms involved in CD44 regulation of SMURF1 and the maintenance of an HNSCC CSC phenotype by comparing the tumorigenic and invasive properties of CD44-high cells to those of CD44 knockout (KO) cells. Treatment of CD44-high cells with HA reduced BMP signaling and increased their migration in a transwell assay. In contrast, CD44-KO cells exhibited reduced SMURF1 protein expression and decreased transwell migration. CD44 KO also greatly reduced colony formation of a highly invasive and recurrent HNSCC cell line, but only partially reduced the clonogenicity of a less-invasive cell line. CD44-high cells also exhibited an invasive and CSC-like growth pattern in organotypic culture (OTC) assays. In contrast, CD44-KO cells failed to generate an epithelial-to-mesenchymal transition (EMT) phenotype in OTC, leading to a loss of invasive capacity. Reconstitution of CD44 expression appears to at least partially restore the invasive and EMT

phenotypes. In summary, we show that although CD44 plays a minor role in cell migration, it may be crucial for maintaining an anchorage-independent growth and invasive phenotype of HNSCC.