
Despite numerous advances in treatment, the 5-year survival rate for head and neck squamous cell carcinoma (HNSCC) has remained largely unchanged for the past 50 years. This poor outcome is due to a number of variables, including the development of multiple primary tumors. Therefore, it is essential to supplement early detection with effective chemopreventive strategies. Using the carcinogenic agent 4-nitroquinoline 1-oxide (4-NQO) to produce HNSCC in a mouse model, we tested the hypothesis that ZD6474 is an effective chemopreventive agent in a preclinical animal model of HNSCC. CBA mice were treated with 4-NQO (100 µg/mL) in their drinking water for a period of 8 weeks. The mice were then randomized to either no treatment or oral lavage of ZD6474 (25 mg/kg/d) for 24 weeks (total time of experiment 32 weeks). At the completion of the study, the proportion of mice with dysplasia or HNSCC was significantly different between the 2 treatment groups (96% in the control and 28% in the ZD6474 group; Fisher exact test: P < .001). Similarly, the proportion of animals with HNSCC was significantly different between the 2 treatment groups (71% in the control and 12% in the ZD6474 group; Fisher exact test: P < .001). In addition, animals treated with ZD6474 displayed a lower proliferative index, a decrease in microvessel density, and the inhibition of phosphorylation of EGFR and VEGFR2 compared with the control mice. These data support the hypothesis that ZD6474 is an effective chemopreventive agent in a preclinical animal model of HNSCC. CBA mice were treated with 4-NQO (100 µg/mL) in their drinking water for a period of 8 weeks. The mice were then randomized to either no treatment or oral lavage of ZD6474 (25 mg/kg/d) for 24 weeks (total time of experiment 32 weeks). At the completion of the study, the proportion of mice with dysplasia or HNSCC was significantly different between the 2 treatment groups (96% in the control and 28% in the ZD6474 group; Fisher exact test: P < .001). Similarly, the proportion of animals with HNSCC was significantly different between the 2 treatment groups (71% in the control and 12% in the ZD6474 group; Fisher exact test: P < .001). In addition, animals treated with ZD6474 displayed a lower proliferative index, a decrease in microvessel density, and the inhibition of phosphorylation of EGFR and VEGFR2 compared with the control mice. These data support the hypothesis that ZD6474 may be promising chemopreventive agent for individuals at risk for developing HNSCC.


Dyskerin is a multifunctional protein that is commonly up-regulated in neoplasia, including oral squamous cell carcinoma (OSCC). Dyskerin binds to and stabilizes noncoding RNAs, including a subset of small nucleolar RNAs (snRNA), which are characterized by a common H/ACA secondary structure. Because dyskerin is a ubiquitous protein, we hypothesized that its loss of function would reduce the accumulation of all H/ACA snoRNA, regardless of cell type. To test this hypothesis, we transfected UM-SCC1 OSCC cells and U2OS osteosarcoma cells with dyskerin small interfering (siRNA); siRNAs directed against GAPDH and a nonspecific target served as negative controls. Forty-eight hours later, total RNA was extracted, reverse transcribed, and subjected to real-time polymerase chain reaction analysis. Loss of dyskerin function significantly reduced the levels of 3 randomly selected H/ACA snoRNA, including U17A, U19, and U66, by more than twofold in both cell lines relative to the controls. In contrast, the levels of 1D box snoRNA, snoRNA USA, and miR-let-7g remained essentially unchanged; dyskerin is not known to bind to these RNA. However, dyskerin was recently shown to directly bind to micro-RNA (miRNA) sequences that are embedded within and processed from H/ACA snoRNA. miRNAs are a class of small noncoding RNA that regulate posttranscriptional gene expression. We have now shown for the first time that loss of dyskerin also significantly decreased the levels of the miRNA miR-664, miR-1248, and miR-1291, as well as their corresponding precursors. The roles of these dyskerin-regulated noncoding RNAs in tumorigenesis are not currently known. Nonetheless, these findings suggest new and novel avenues of investigation into the molecular mechanisms by which dyskerin may contribute to neoplasia.


Spindle cell squamous cell carcinoma (SCSCC) is a rare bimorphic malignant neoplasm of the upper aerodigestive tract and skin. This tumor is composed of a squamous cell carcinoma (SCC) and a malignant spindle component; therefore, diagnosis on hematoxylin and eosin–stained sections alone presents a challenge. The present case had serendipitous findings of tissue-bound and serum autoantibodies on direct and indirect immunofluorescence (IF), respectively.

Objectives. The aim of this study was to report the immunohistochemistry (IHC) findings in a case of oral SCSCC and to further investigate the nature of the patient’s circulating autoantibodies by using an enzyme-linked immunosorbent assay (ELISA).

Results. IHC showed that the carcinomatous portion of the tumor expressed pankeratin, epithelial membrane antigen, and E-cadherin; the sarcomatoid component expressed vimentin, smooth muscle antigen, and N-cadherin; both carcinomatous and sarcomatoid portions of the tumor expressed p63 and CK34BE12. Direct and indirect IF showed autoantibodies in a stratified epithelial specific-antinuclear antibody pattern; ELISA
showed that these patient antibodies were of the IgG class and were immunoreactive with recombinantly produced delta-Np63.

**Conclusions.** Studies have shown that overexpression of the p63 gene is reported in 85% of oral SCC cases and that overexpressed p63 functions as an oncogene. This case shows that p63 is useful as an IHC marker of both the squamous and the sarcomatoid phenotypes in SCSSC. In addition, we present evidence of a humoral immune response to the overexpressed p63 protein. Autoantibodies to p63 were previously demonstrated in chronic ulcerative stomatitis (CUS) and lichen planus (LP). This case raises the question of whether p63 autoantibodies may represent potentially malignant cases of CUS or LP. Ongoing studies are exploring the role of p63 autoantibodies as SCC biomarkers.

**PITFALLS IN DIAGNOSTIC TESTS FOR HIGH-RISK HUMAN PAPILLOMAVIRUS.** P. DeVilliers, A. Andea, E. Kerr, L. Novak. U Alabama, Birmingham.

High-risk human papillomavirus (HR-HPV) is associated with some cases of oral and pharyngeal invasive squamous cell carcinoma (OPSCC). The preferred methods for detecting HR-HPV are in situ hybridization (ISH) and polymerase chain reaction (PCR). There are conflicting reports on the reliability of immunohistochemistry (IHC) detection of HPV types 6, 11, 16, 18, 31, 33, 42, 51, 52, 56, and 58 as well as the significance of p16 as a surrogate marker of HR-HPV status. This study evaluated the potential utility of combining IHC for HPV and p16 as less expensive alternative testing methods to ISH or PCR for detecting high-risk HPV. Immunohistochemical expression of p16 (MTM Laboratories, Westborough, MA) and HPV (Dako, Denmark) was analyzed in tissue blocks from patients recently diagnosed with oral and pharyngeal epithelial dysplasia or invasive squamous cell carcinoma with confirmed HPV status by ISH and PCR. Cases were evaluated for distribution, extent, and degree of intensity of p16 and HPV IHC stain. Cases of OPSCC that were originally diagnosed as HR-HPV positive by PCR were also positive by ISH and were strongly positive for p16. Cases of squamous papilloma and dysplasia that were diagnosed as low-risk HPV positive by PCR showed only focal staining for p16. The degree of p16 expression correlated with the severity of dysplasia. Cases of OPSCC with negative HR-HPV expression by PCR/ISH were negative for p16. All cases were HPV negative by immunohistochemistry. These data imply that: 1) immunohistochemical expression of p16 is a reliable surrogate marker for HR-HPV expression in OPSCC; 2) HR-HPV detection by ISH is a reliable alternative when PCR testing is not available; and 3) HPV detection by IHC is not dependable.

**THE ROLE OF DIRECT VISUAL FLUORESCENT EXAMINATION (VELSCOPE) IN TUMOR MARGIN DELINEATION AND ROUTINE SCREENING FOR ORAL CANCER.** K. McNamara, A. Agrawal, T. Teknos, E. Ozer, E. Evans, C. Allen, J. Kalmar. Ohio State U, Columbus.

Velscope is a commercially available oral cancer screening system based on principles of tissue fluorescence. It has been proposed that direct visual fluorescent examination (DVFE) of the oral cavity may be a useful adjunct to conventional oral examination (COE), however, evidence for this role is lacking. In addition, DVFE reportedly improves tumor margin delineation in the operating room. We present initial results from a 2-arm study to evaluate DVFE in both surgical margin delineation and routine screening of the oral cavity. Twenty patients presenting for surgical excision of prior biopsy–confirmed oral epithelial dysplasia or squamous cell carcinoma were included in the high risk study arm. Lesional tissue margins were assessed by both COE and DVFE, and punch biopsy was used to provide histopathologic correlation. A total of 33.3% of tumors exhibited positive DVFE extension beyond the clinically visible tumor margin, and 58.3% of these extensions demonstrated microscopic evidence of premalignancy (sensitivity 64%, specificity 62%). The general population study arm consisted of 40 patients presenting for routine dental care. Study subjects received a comprehensive COE followed by DVFE, and all positive DVFE areas were referred for scalpel biopsy. DVFE positivity was not significantly related to biopsy evidence of cancer or precancer (P = .0016). These results confirm that DVFE may be useful in tumor margin delineation during surgical management of patients with oral (pre)cancer. False-positive test results, however, may limit the utility of Velscope as a routine screening device.


The diagnosis of ameloblastoma or keratocystic odontogenic tumor (odontogenic keratocyst) is usually a routine matter for the practicing oral and maxillofacial pathologist owing to well-established microscopic criteria. There are a number of cases reported as keratoameloblastoma (KAB) or solid keratocystic odontogenic tumor (SKOT) where this distinction is not so clear. Fewer than 15 cases of KAB and 3 cases of SKOT have been documented in the literature. This is a report of a case that demonstrates the dilemma that a pathologist may face in distinguishing between these 2 rare microscopic presentations of 2 of the most common odontogenic neoplasms. During a routine dental examination, a 25-year-old woman was discovered to have an asymptomatic 1.0-cm ovoid radiolucency in the right mandibular body between the roots of vital teeth #28 and #29. The lesion was biopsied, and a microscopic diagnosis of "odontogenic keratocyst, solid variant" was rendered. Microscopic criteria as well as the clinical significance for the diagnosis of KAB and SKOT are discussed.


First described by Pindborg in 1955, the calcifying epithelial odontogenic tumor (CEOT) is a benign locally invasive neoplasm of odontogenic epithelium without odontogenic ectomesenchyme. This lesion is uncommon, with only ~200 cases reported in the literature. Radiographically, CEOTs present as unic- or multilocular mixed lucent/opaque lesions of varying size. CEOTs typically appear as solitary lesions. In rare instances, multiple lesions may occur within the same patient. We report the case of a 39-year-old Hispanic woman presenting with bilateral mandibular mixed radiolucent/radiopaque lesions associated with 3 impacted molars. Bilateral incisional biopsies revealed findings that supported the diagnosis of CEOT, including islands, nests, and cords of epithelial cells within a fibrous to myxoid stroma; the presence of amyloid-like material; and Liesegang ring calcifications. This unique case demonstrates the ability of CEOTs to