Angiogenic Squamous Dysplasia in Oral Epithelial Dysplastic Lesions

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Abstract: Statement of problem: Angiogenic squamous dysplasia (ASD), a qualitative distinct form of angiogenesis, was first described in pre-invasive bronchial mucosa of high-risk individuals. It is characterized histologically by the presence of capillary tufts that are closely juxtaposed to and projecting into the dysplastic bronchial epithelium. Objective: To determine whether ASD occurs in oral epithelial dysplastic lesions. Methods: Sixty cases of potentially malignant oral epithelial lesions comprising 20 mild epithelial dysplasia (ED), 20 moderate ED and 20 severe ED(inclusive of carcinoma-in-situ), and 10 normal oral mucosa (as normal controls) were retrieved from the archives of the Department of Oral Pathology, Oral Medicine & Periodontology, Faculty of Dentistry, University of Malaya, and the Cancer Research Centre, Institute for Medical Research, Kuala Lumpur, Malaysia. The grading of oral ED was in accordance with the recommendations of the World Health Organization. For all selected cases, new 5 micron thick sections were prepared for H&E staining, and for immunohistochemistry with three vascular markers, CD31, CD34 and CD105, with appropriate positive and negative controls. The established criteria for identification of ASD was applied. Results: ASD was identified in parts of severe oral ED, but was absent in normal oral epithelium, mild and moderate oral ED. About 65% of these severe oral ED cases were from individuals with high-risk habits. As with bronchial ASD, the capillaries typically formed CD31- and CD34-positive projections or loops that abut onto the overlying dysplastic oral epithelium causing the latter to assume a papillary surface configuration. Unlike bronchial ASD which occurs in respiratory-type epithelium, oral ASD was found in keratinzed stratified squamous epithelium. CD105 confirmed the presence of neoangiogenesis. Conclusions: Present findings confirm that ASD can occur in oral severe ED. It also demonstrates that this angiogenic abnormality is not unique to bronchial mucosal dysplastic lesions.

Keywords: Oral epithelial dysplasia, angiogenesis, immunohistochemistry

Introduction

Oral potentially malignant epithelial lesions(PMELs) are defined as morphologically altered tissues in which cancer is more likely to occur than in its apparently normal counterpart. It indicates all the stages on the route to oral malignancy.

Angiogenesis is defined as the growth and proliferation of blood vessels from the existing vasculature. It is a multistep process that is mediated by vascular endothelial cells and involves the response of existing vasculature to a series of protein mediators. Angiogenic squamous dysplasia (ASD), a qualitative distinct form of angiogenesis, was first described in pre-invasive bronchial mucosa of high-risk individuals. It is characterized histologically by the presence of capillary tufts that are closely juxtaposed to and projecting into the dysplastic bronchial epithelium.[1-4)]

Various endothelial markers such as CD31, CD34 and CD105 are used in immunohistochemistry to determine angiogenic activity. CD31 and CD34 are pan-endothelial markers whereas CD105 is a marker for neoangiogenesis.

The aim of this study was to determine whether ASD occurs in oral PMELs,and to define their histopathological characteristics if these lesions are encountered.

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Materials and Methods

Sample:

Sixty cases of oral PMELs comprising 20 mild epithelial dysplasia (ED), 20 moderate ED and 20 severe ED (inclusive of carcinoma-in-situ), and 10 normal oral mucosa (as normal controls) were retrieved from the archives of the Department of Oral Pathology, Oral Medicine & Periodontology, Faculty of Dentistry, University of Malaya, and the Cancer Research Centre, Institute for Medical Research, Kuala Lumpur, Malaysia. The grading of oral ED was in accordance with WHO recommendations. For all selected cases, serial 5ìm thick sections were prepared for H&E staining, and labeled in a consecutive order.

Immunohistochemical staining:

Immunohistochemical staining with CD31, CD34 and CD105 were performed using the avidin biotinylated complex system (ABComplex/HRP, DAKO) and with appropriate positive and negative controls.

ASD identification:

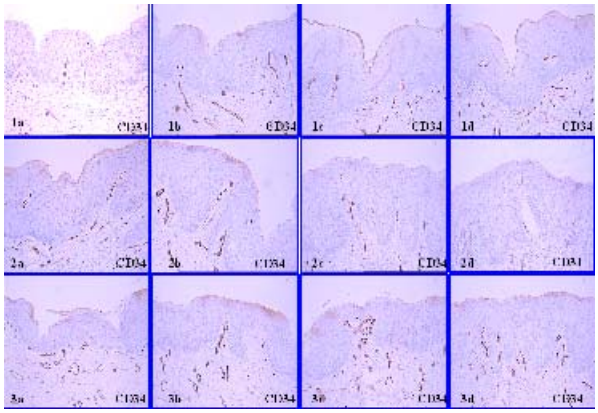
The established criteria for the identification of ASD was applied. [1-4)] Slides were evaluated at low and medium power (x4 and x20 objectives) to determine areas in the oral mucosa presenting with ASD lesions. Serial images of these lesions were digitally captured to determine their 3-D microcapillary patterns.

Results and Discussion

ASD was identified in parts of severe oral ED, but was absent in normal oral epithelium, mild and moderate oral ED. About 65% of these severe oral ED cases were from individuals with high-risk habits. As with bronchial ASD, the capillaries typically formed CD31- and CD34-positive projections or loops that abut onto the overlying dysplastic oral epithelium causing the latter to assume a papillary surface configuration (Figs. 1-3). Unlike bronchial ASD which occurs in respiratory-type epithelium, oral ASD was found in keratinized stratified squamous epithelium. CD105 confirmed the presence of neoangiogenesis.

The close juxtaposition of capillary tufts to the dysplastic epithelium suggests that an angiogenic stimulus may be associated with epithelial dysplasia in the upper aerodigestive tract.These angiopapillary changes are most likely a consequence of architectural rearrangement of the capillary microvasculature within the oral dysplastic epithelium.
Possible mechanisms of angiogenesis in ASD may involve small populations of dysplastic squamous cells harboring premalignant mutations transmitting angiogenic signals over very short distance. [1)]

We concluded that ASD:
- is not unique to bronchial pre-invasive mucosa
- of the oral mucosa may represent an important intermediate pathological biomarker preceding oral cancer development,
- may therefore be useful as a predictive marker of malignancy.
- could have significance in relation to the treatment of these lesions.



Figs1-3. – Photomicrographs showing severe oral epithelial dysplasia exhibiting capillaries that typically formed CD31- and CD34-positive projections or loops. These small blood vessels abut onto the overlying dysplastic oral epithelium causing the latter to assume a papillary surface configuration.

Table 1. Histopathology of oral angiogenic squamous dysplasia

No.	Key findings
1.	SSE shows severe dysplasia with hypercellularity, cellular pleomorphism and incomplete maturation.
2.	Invasion of papillary connective tissues by collections of capillary –sized blood vessels.
3.	Projection of these capillaries onto the overlying dysplastic epithelium, thus imparting a surface papillary configuration.
4.	Serial sections of these capillary projections confirmed that they represent intramucosal capilllary loops

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