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A novel histopathological subclassification of oral submucous fibrosis based on epithelial keratinization: A proof-of-concept pilot observation

Gargi S. Sarode, Sachin C. Sarode *

Department of Oral Pathology and Microbiology, Dr. D.Y. Patil Dental College and Hospital, Dr. D.Y. Patil Vidyapeeth, Sant-Tukaram Nagar, Pimpri, Pune, 411018, MH, India

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ABSTRACT

Oral submucous fibrosis (OSMF) is a chronic, progressive oral potentially malignant disorder strongly associated with areca nut consumption. Despite extensive research, a reliable and clinically applicable biomarker for predicting malignant transformation remains unavailable. The present pilot investigation introduces a novel concept by classifying OSMF into two histopathological variants—hyperkeratinized and non-hyperkeratinized—based on epithelial keratinization patterns, a distinction not previously described in the literature. Archival biopsy samples of 51 histologically confirmed OSMF cases from the buccal mucosa were analyzed for keratinization type and epithelial dysplasia. Additionally, 37 cases of oral squamous cell carcinoma arising in the background of OSMF (OSCC-OSMF) were examined for keratinization at surgical margins. Among OSMF cases, 19.6 % exhibited hyperkeratinization, all showing high-risk epithelial dysplasia, whereas 90.2 % of normally keratinized cases demonstrated low-risk dysplasia. In OSCC-OSMF specimens, 60 % of surgical margins revealed hyperkeratinization. These findings suggest that epithelial hyperkeratinization may represent an early histological alteration associated with malignant transformation rather than a protective epithelial response. Although limited by small sample size, this study provides proof-of-concept for a novel histopathological subclassification of OSMF, warranting validation through larger, multicentre longitudinal and molecular correlation studies.

1. Introduction

Oral submucous fibrosis (OSMF) is a prevalent oral potentially malignant disorder, especially in Asian countries caused by chromic consumption of betel quid (also called Gutkha).[1] The pathogenesis involves increased fibrosis in the underlying connective tissue stroma, contributing to fibrosis-mediated oncological changes and transformation of oral squamous cell carcinoma (OSCC).[2] According to a recent systematic review, the malignant transformation rate of OSMF is 4.2 %, which is significant enough to warrant investigations on finding appropriate markers for predicting malignant potential as well as early detection of OSCC.[3] Various attempts have been made in the past to investigate clinical, histopathological, biochemical, and molecular markers for predicting the malignant potential of OSMF, unfortunately no reliable and clinically practicable marker emerges till date.[4]

Considering the limited resources in low- and middle-income countries, where OSMF is most prevalent, histopathological features are the most feasible, routinely performed and affordable method for investigating malignant potential through assessing the epithelial dysplasia. However, assessment of epithelia dysplasia becomes very challenging in OSMF tissues due to atrophic epithelium and loss of rete rides which makes the distinction of epithelial layers strata impossible. Although attempts to identify the unique dysplastic features in the atrophic epithelium has been made in the literature,[5] more and more attempts into this niche area is the need of the hour. This present investigation, we discusses our observations on histopathological parameters in OSMF cases, which warrant special attention and in-depth investigation in the future.

2. Methods and observations

The present observation is based on the histological examination of 51 OSMF cases from the archives of the department. The study represents lateral observations made on the protocol approved by the institutional ethics committee (DYPDCH/IEC/120/72/19). As this study was designed as a pilot, proof-of-concept observation without comparative

* Corresponding author. Department of Oral Pathology and Microbiology, Dr. D. Y. Patil Dental College and Hospital, Dr. D.Y. Patil Vidyapeeth, Sant-Tukaram Nagar, Pimpri, Pune, 18, Maharashtra, India.
E-mail address: drsachinsarode@gmail.com (S.C. Sarode).

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groups, no inferential statistical analyses were performed; instead, descriptive proportions were used to summarize the distribution of keratinization patterns across cases. All biopsy samples location was from the buccal mucosa, which is biologically and histopathologically considered non-keratinized or sometimes parakeratinized in healthy individuals. Diagnostic criteria proposed by the Pindborg et al. was used to diagnose OSMF cases.[6] Although, keratinization feature and type is very basic skills, the slides were investigated by two pathologies [SS and SG] and no disagreement on the diagnosis was reported. We observed that some cases showed hyperkeratinization, while others did not show any keratinization or parakeratinization. To simplify the distinction, we divided the cases into hyperkeratinized and normally keratinized (non-keratinized or parakeratinization). Of the 51 cases studied, 10 (19.6 %) were categorized as hyperkeratinized, and 41 (80.39 %) as normal keratinized [26 (50.98 %) with parakeratinization and 15 (29.41 %) with non-keratinized mucosa] (Fig. 1). Interestingly, all cases with hyperkeratinized epithelium showed high-risk epithelial dysplasia, while 37 (90.24 %) cases of normal keratinized OSMF showed low-risk epithelial dysplasia, with four cases of non-dysplastic features. A literature search revealed similar characterizations of OSMF cases based on keratinization. In their study of 30 OSMF patients, Caniff et al.[7] observed that 33 % of patients had non-keratinized or poorly keratinized epithelium, 67 % had keratinized metaplasia, 13 % had parakeratinization, and 23 % had hyper-orthokeratinization.

A pragmatic question can be raised: would such a distinction help predict malignant potential in OSMF patients? To address this query, we further investigated the histopathology features of the surgical margins associated with excised specimens of OSCC which occurred against a background of OSMF (OSCC-OSMF). Based on the previous attempts in the literature,[5] we believed that the epithelial status of surgical margins in OSCC-OSMF displays all features relevant to malignant transformation. The diagnostic criterial for inclusion of inclusion of OSCC-OSMF cases were adopted from our previous publication.[5] We investigated 37 cases of OSCC-OSMF and identified 15 cases displaying the buccal mucosa as one of the surgical margins. Out of these 15 cases, 9 (60 %) showed hyperkeratinization, while 6 (40 %) were non-hyperkeratinized [1 (6.66 %) parakeratinized and 5 (33.3 %) non-keratinized mucosa]. The high number of OSCC-OSMF cases showing hyperkeratinization at the surgical margins suggests a potential prognostic marker for malignant transformation.

3. Discussion and conclusion

The present observation clearly demonstrates two different types of OSMF based on keratinization status, i.e., hyperkeratinized and normal keratinized with hyperkeratized mucosa most frequently noted at surgical margins of OSCC-OSMF cases. This suggest potentiality of this feature as future marker of deeper investigation. In support of this contention, Vaidya et al. has reported distinctive cytokeratin patterns in different oral potentially malignant disorders of the oral cavity.[8]

Keratinization is often seen in patients who consumes smokeless tobacco. Whether this variation in keratinization in OSMF depends on the tobacco content in betel quid or Gutkha or represents an exaggerated defence mechanism of epithelium is a question that needs further exploration. This requires an well standardized in-depth investigation into the chewing habits of OSMF patients and its correlation with histopathology of buccal mucosa tissues.

One school of thought considers hyperkeratinization a physical barrier and a type of body defence mechanism that protects the underlying epithelial cells from harmful carcinogens.[9] If this contention holds true, it contradicts the present observation of more incidence of hyperkeratinization in OSCC-OSMF surgical margins specimens and also questions it as a potential marker. Nonetheless, this demands more in depth investigation on the molecular mechanisms involved in the

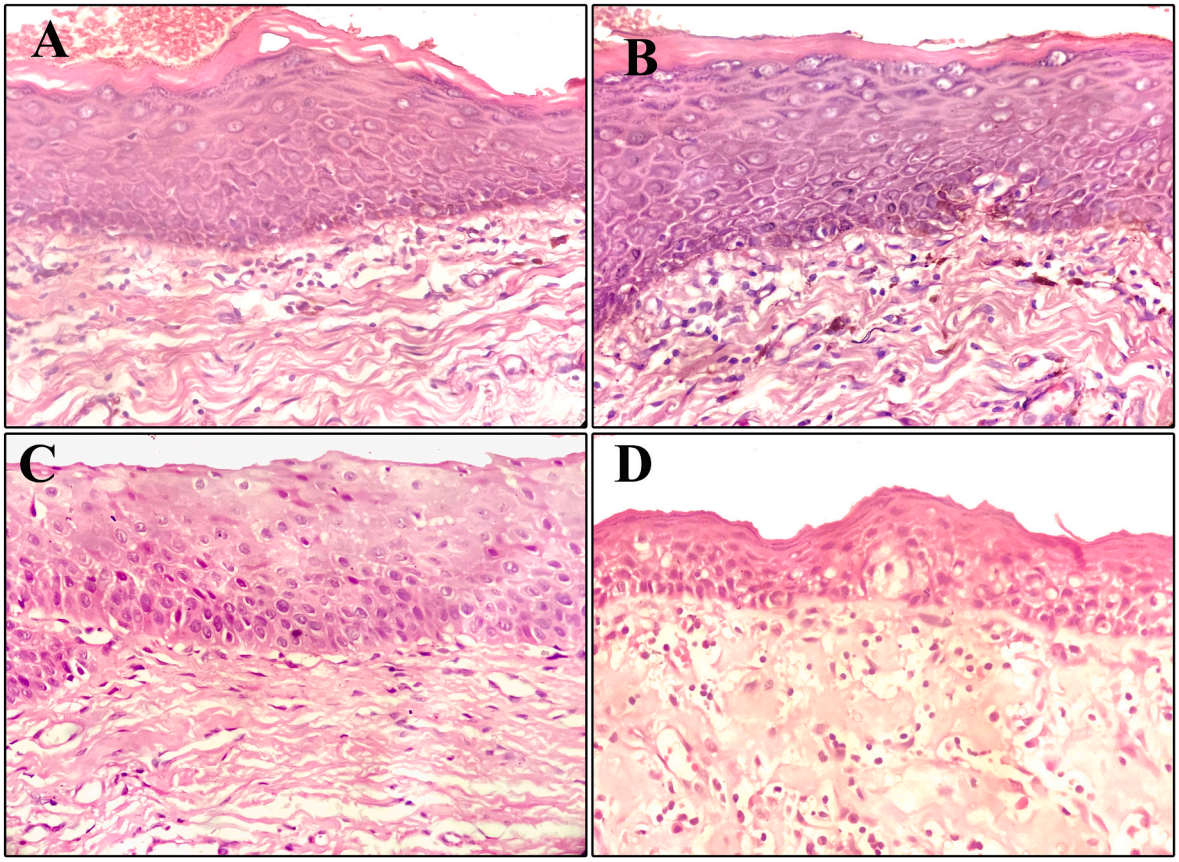

Fig. 1. Photomicrograph of oral submucous fibrosis showing two distinct variants: hyperkeratinized (A and B) and non-hyperkeratinized (C and D).

hyperkeratinization in OSMF patients.

Although the limited sample size is the major limitation of the present pilot investigation, and our conclusions are based on observations made at surgical margins in OSCC-OSMF specimens (which is not an ideal disease model for confirming malignant potential), this approach provides valuable proof-of-concept data for future work. Due to the pilot nature and small archival dataset, advanced statistical or reliability analyses were not feasible and the findings are presented as preliminary, hypothesis-generating observations. Future validation should ideally employ a multicentre prospective cohort with longitudinal follow-up, a nested case–control design for molecular correlation, and an external retrospective validation cohort to comprehensively establish the prognostic utility of hyperkeratinization as a biomarker of malignant transformation.

As far as histopathology is concerned, studies in the literature have mainly focused on the epithelial dysplastic features and fibrosis in OSMF patients. We found only one study describing keratinization profile in detail.[7] Based on the preliminary observations of the present study, we have generated a proof of concept for possibly dividing OSMF into hyperkeratinized and non-hyperkeratinized variants, which deserves further investigation in future studies. The more frequent presence of dysplastic features in hyperkeratinized OSMF and at surgical margins of OSCC-OSMF further supports the potential of this proposition. We recommend future studies using a cohort study design with larger samples and good follow-up to prove the proposed hypothesis. Such distinction might impact clinical decision-making, patient management, and follow-up strategies. Taken together, these findings should be interpreted strictly within the constraints of a small, retrospective pilot dataset, and are intended to serve only as preliminary observations that warrant validation through larger and better-designed cohort studies.

Patient's/Guardian's consent

This is retrospective study on archival formalin fixed paraffin embedded tissues. The institution has obtained blanket consent form the patient for the use of biological material for research purpose.

Ethics statement

The investigations presented in the present study represents lateral observations made on the protocol approved by the institutional ethics committee (DYPDCH/IEC/120/72/19).

Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this work the authors used ChatGPT 5 in order to improve the English language and readability of the manuscript. After using this tool/service, the authors reviewed and edited the content as needed and takes full responsibility for the content of the published article.

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Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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References

1. Kondaiah P, Pant I, Khan I. Molecular pathways regulated by areca nut in the etiopathogenesis of oral submucous fibrosis. *Periodontol 2000*. 2019 Jun;80(1):213–224. https://doi.org/10.1111/prd.12266.
2. Yuwanati M, Ramadoss R, Kudo Y, Ramani P, Senthil Murugan M. Prevalence of oral submucous fibrosis among areca nut chewers: a systematic review and meta-analysis. *Oral Dis*. 2023 Jul;29(5):1920–1926. https://doi.org/10.1111/odi.14235.
3. Kujan O, Mello FW, Warnakulasuriya S. Malignant transformation of oral submucous fibrosis: a systematic review and meta-analysis. *Oral Dis*. 2021 Nov;27(8):1936–1946. https://doi.org/10.1111/odi.13727.
4. Lin F, Xiao T, Wang B, et al. Mechanisms and markers of malignant transformation of oral submucous fibrosis. *Heliyon*. 2023 Dec 7;10(1), e23314. https://doi.org/10.1016/j.heliyon.2023.e23314.
5. Sarode SC, Chaudhary M, Gadbail A, Tekade S, Patil S, Sarode GS. Dysplastic features relevant to malignant transformation in atrophic epithelium of oral submucous fibrosis: a preliminary study. *J Oral Pathol Med*. 2018 Apr;47(4):410–416. https://doi.org/10.1111/jop.12699.
6. Pindborg JJ, Murti PR, Bhonsle RB, Gupta PC, Daftary DK, Mehta FS. Oral submucous fibrosis as a precancerous condition. *Scand J Dent Res*. 1984;92(3):224–229. https://doi.org/10.1111/j.1600-0722.1984.tb00883.x.
7. Canniff J, Harvey W, Harris M. Oral submucous fibrosis: its pathogenesis and management. *Br Dent J*. 1986;160:429–434. https://doi.org/10.1038/sj.bdj.4805876.
8. Vaidya MM, Sawant SS, Borges AM, Ogale SB, Bhisey AN. Cytokeratin expression in precancerous lesions of the human oral cavity. *Oral Oncol*. 1998;34(4):261–264.
9. Payne TF. Why are white lesions white? Observations on keratin. *Oral Surg Oral Med Oral Pathol*. 1975 Nov;40(5):652–658. https://doi.org/10.1016/0030-4220(75)90375-8.