The Complexity of the Oral Mucosa: A Review of the Use of Fractal Geometry

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Abstract: The oral mucosa represents the lining of the oral cavity. It is composed of epithelium supported by connective tissue. The shape of the interface between these two tissues has traditionally been used to describe physiological and pathological changes in the oral mucosa. The aim of this study was to review the use of fractal geometry to compare the complexity of the epithelial connective tissue interface (ECTI) in normal, dysplastic, pseudo-neoplastic and malignant oral mucosae. This was achieved by estimating global and local fractal dimensions of ECTI profiles isolated from histological images representing these different diagnostic groups. This study showed that fractal geometry is a useful objective tool in describing tissue complexity in the oral mucosa.

Keywords: oral mucosa, squamous cell carcinoma, epithelial dysplasia, pseudoepitheliomatous hyperplasia

1. INTRODUCTION

The oral mucosa represents the lining of the oral cavity. It is composed of stratified squamous epithelium supported by a connective tissue called the lamina propria. Both tissues are attached to each other via a basal lamina.

The shape and regularity of the epithelial connective tissue interface (ECTI), has classically been used to describe both physiological and pathological changes in the oral mucosa. However, the description of ECTI irregularity has most often been subjective. The use of fractal geometry has therefore been introduced to describe the irregularity of ECTI profiles taken from the oral mucosa both in normal and in abnormal cases.

1.1. A Few Definitions

Epithelial dysplasia is a term used to describe tissue changes in lesions that have a higher risk of undergoing malignant transformation than their normal counterparts. Classically, it is graded (subjectively) according to the extent of pathological changes into mild, moderate and severe dysplasia.

Squamous cell carcinoma (SCC) is the most common malignant tumour of the oral cavity. It is a malignant neoplasm of the stratified squamous epithelial lining of the oral cavity.

Pseudo-epitheliomatous hyperplasia (PEH) is a reactive, non-neoplastic proliferation of epithelium seen in association with some neoplasms, infections and inflammatory processes.

The epithelium in PEH is characterised by a hyperplastic change that mimics morphologically the invasion patterns of SCC.

1.2 Aim

The aim of this study was to review the use of fractal geometry to compare the tissue complexity in normal, dysplastic, pseudo-neoplastic and malignant oral mucosae. This was achieved by estimating both the global and local fractal dimensions of ECTI profiles isolated from histological sections representing these different entities.

2. MATERIAL AND METHODS

Histological images were captured at x4 magnification. Image analysis was then performed to separate the epithelium from the connective tissue using multiple thresholding (Otsu, 1979). The ECTI profiles were then extracted using a Laplacian edge detecting convolution filter (Landini et al., 2000) (Figure 1).

The irregularity of these profiles was subsequently estimated using fractal geometry principles. The box counting method was used to estimate the global complexity (Landini, 1996), while the mass radius method (ranging sizes from 47µm-557µm) was used to estimate the local complexity (Vicsek, 1996). Seven different box sizes for the local and local connected fractal dimensions estimation were used to depict any differences between diverse diagnostic entities at different scales.

3. RESULTS AND DISCUSSION

3.1 Normal Oral Mucosa at Different Sites in the Oral Cavity (Abu Eid and Landini, 2003)

The fractal dimension of 75 ECTI profiles representing 21 sections from normal oral mucosa were analysed. These included 10 cases from the buccal mucosa, 4 from the lingual mucosa and 7 from the palatal mucosa.

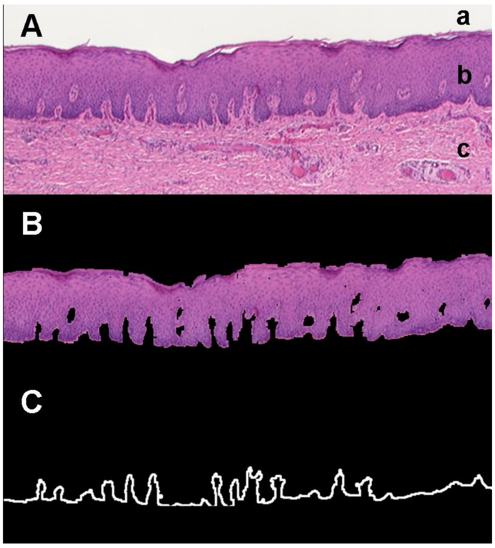
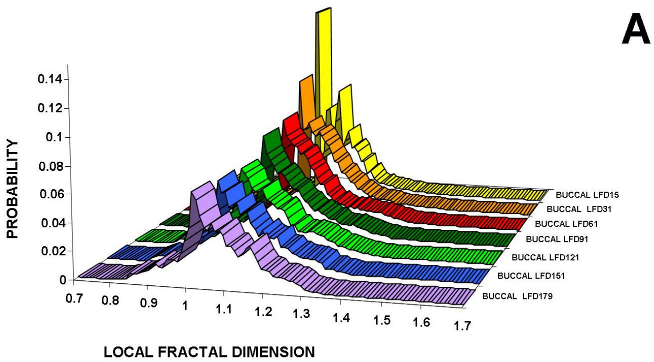
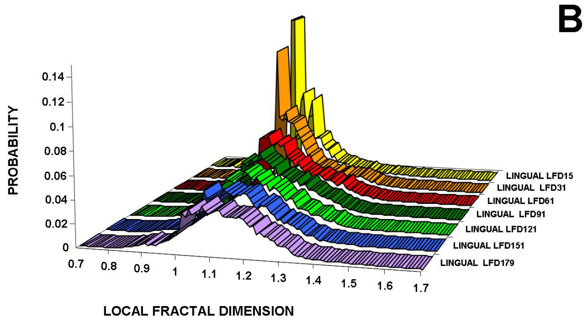


Fig. 1. A) Haematoxylin and Eosin stained normal oral mucosa section x4 (field width 716 pixels). (a) represents the background, (b) represents the epithelium and (c) represents the connective tissue. B) The isolated epithelium of the epithelium in A. C) ECTI of the epithelium in A.

The mean box fractal dimensions showed that the palatal mucosa was the most complex (D = 1.16 + 0.05), followed by (though not statistically different) the lingual mucosa (D = 1.13 + 0.08) and the buccal mucosa had the lowest fractal dimension (D = 1.07 + 0.06), and was found to be statistically different from the palatal and the lingual mucosae.

The distribution of both local and local connected fractal dimensions also showed that the ECTI profiles in the palatal mucosa were more complex than those in the lingual mucosa, which in turn were more irregular than ECTI profiles in the buccal mucosa. This was particularly apparent with large window sizes (190µm -557µm). However with the smaller window sizes (47µm -96µm) the local and local connected fractal dimension distributions were similar in different groups (Figure 2). These results agree with the observations of Bale and White (1982) (Bale and White, 1982), who described more prominent rete ridges in the palatal mucosa than the buccal mucosa which are thought to enable the mucosa to withstand high mechanical forces.





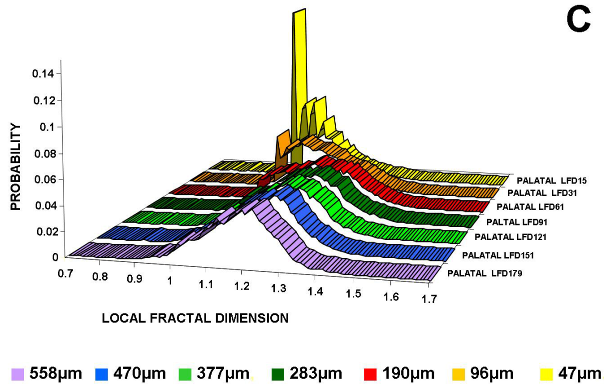


Fig. 2. Histograms of the local fractal dimensions distribution at the different window sizes for: A) Normal Buccal Mucosa. B) Normal Lingual Mucosa. C) Normal Palatal Mucosa

3.2 Changes in ECTI Irregularity in Normal, Dysplastic and Neoplastic Profiles (Abu Eid and Landini, 2003)

Three hundred and fourteen ECTI profiles from 69 cases (21 normal, 4 mild dysplasia, 15 moderate dysplasia, 13 severe dysplasia and 16 SCC) were analyzed. The normal oral mucosa had the lowest mean box fractal dimension (1.09), followed by dysplastic ECTI(s) with 1.14, 1.16, 1.16 values for mild, moderate and severe dysplasia classes respectively. SCC possessed the highest mean fractal dimension (1.23) (Figure 3).

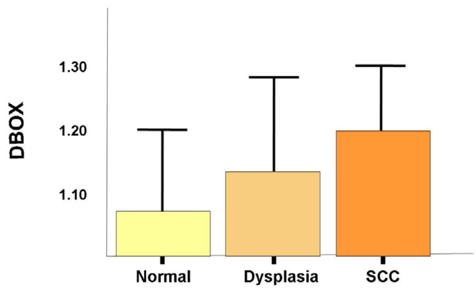


Fig. 3. Mean box fractal dimension (DBOX) of Normal, Dysplasia and SCC ECTI profiles. Vertical bars represent 1 standard deviation.

The distribution of both local and local connected fractal dimensions showed gradual increase from normal, dysplastic to the neoplastic ECTI profiles (Figure 4). The most discriminating results for differentiating the diagnostic classes were achieved using local sets within large window sizes (377µm- 557µm). Up to 94% of normal and SCC profiles, 71% of normal and dysplastic profiles and 70% of dysplastic and neoplstic profiles were correctly classified based on the local complexity of ECTI profiles (hierarchical discriminant analysis with leave-one-out). These findings agree with previous work of Landini and Rippin (1993; 1996) (Landini and Rippin, 1993, 1996).

The discrimination between dysplasia and SCC cases decreased as the severity of dysplasia increased. This was an expected result that reflects the increase in the severity of pathological changes as the grade of dysplasia increases from mild to severe.

The changes in the irregularity of ECTI profiles agree with known mechanism of tumour growth, in which many factors seem to be involved. Defects in the basement membrane and loss of intercellular adhesion may be responsible and contribute to the increased irregularity of tumour margins. Defects in the basement membrane have been observed in many invasive carcinomas including oral carcinomas (Niimi et al., 2001). However, the mechanism of basement membrane breakdown is complicated, due to the involvement of the various components. Oral carcinomas are not the only tumours to be quantitatively described by fractal geometry Fractals have proved useful in describing the complexity of several tumours in various sites of the body such as colorectal polyps (Cross et al., 1994), gallbladder adenocarcinoma (Waliszewski, 1999), basal cell carcinoma of the skin (Miracco et al., 1998) and malignant melanoma (Claridge et al., 1992).

3.3 Dysplasia Grades (Abu Eid and Landini, 2003)

Out of 183 ECTI profiles from 4 mild, 15 moderate and 13 severe dysplasia cases the mean box fractal dimension of the ECTIs were 1.14, 1.16 and 1.16 for mild, moderate and severe dysplasia ECTI profiles respectively (Figure 5). Although there was an increase in the fractal dimension value with increased severity of the dysplasia, those differences were not statistically significant. Even at the largest window size, the local and local connected fractal dimensions failed to differentiate the three dysplasia grades from each others (Figure 6). The worst discrimination (53.2%) (hierarchical discriminant analysis with leave-one-out) was between mild and moderate dysplasia. This suggests that the irregularity of the ECTI may not be correlated to other morphological variables on which the dysplasia grading is based.

Unfortunately, the gold standard used for grading oral epithelial dysplasia is subjective (Pindborg et al., 1985, Landini and Rippin, 1996, Landini and Geake, 1997) and so quantitative methods are now being sought to render diagnosis a more objective activity.

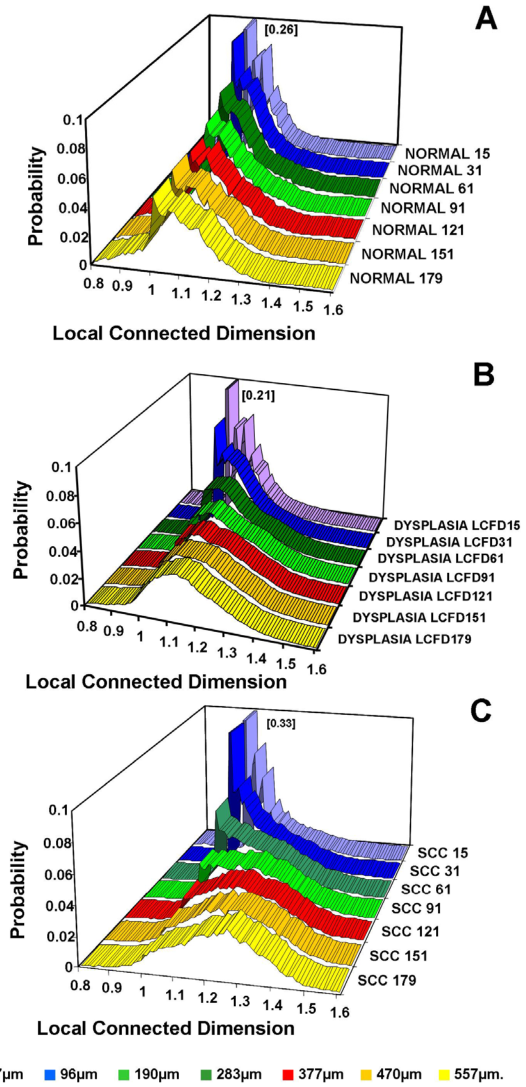


Fig. 4. Histograms of the local connected fractal dimensions distribution at the different window sizes: A) Normal Oral Mucosa. B) Epithelial Dysplasia. C) Squamous Cell Carcinoma. (The highest peaks were truncated off scale for display purposes; the numbers in square brackets indicate the original value at the peak).

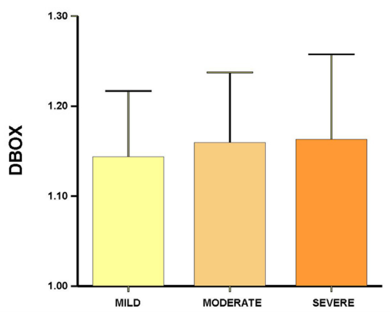


Fig. 5. Mean box fractal dimension (DBOX) of Mild, Moderate and Severe Epithelial Dysplasia. Vertical bars represent 1 standard deviation.

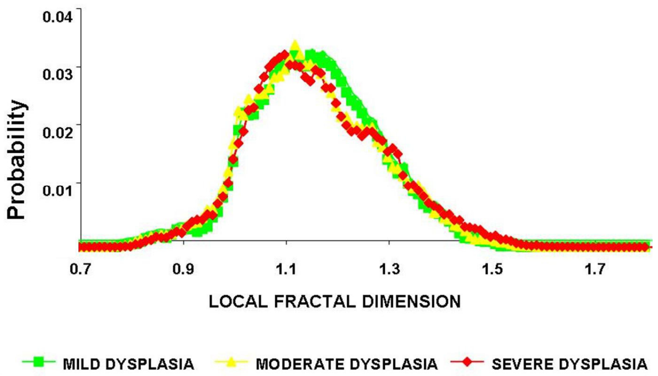


Fig. 6. Local fractal dimension probability of the ECTI for different grades of dysplasia at window size 179 pixels (557µm).

3.4 Pseudoepitheliomatous Hyperplasia, Normal and Dysplastic Mucosae (Abu Eid and Landini, 2005)

The fractal dimensions of 85 ECTI profiles from 12 PEH associated with granular cell tumour (PEH-GCT), 19 epithelial dysplasia and 8 normal oral mucosa cases were calculated. The mean box fractal dimension of the ECTI profiles was 1.28 for GCT-PEH, 1.12 for dysplasia and 1.06 for normal profiles.

Using large window sizes (190µm- 557µm), the distribution of local and local connected fractal dimensions showed that the ECTI complexity was greater in GCT-PEH profiles than in dysplasia cases which in turn proved to be more complex than normal ECTI profiles. With the smaller window sizes (47µm -96µm) the local and local connected fractal dimension distributions were similar in all three groups. Up to 96.4% (hierarchical discriminant analysis with leave-one-out) of the GCT-PEH and normal profiles were correctly classified using the fractal descriptors and up to 94.7% of dysplasia ECTIs (moderate and severe) could be discriminated from PEH profiles.

According to these results, PEH profiles were shown to be more irregular than both normal and dysplastic ECTI profiles. Further studies are needed to characterize the behaviour of PEH-GCT.

3.5 Squamous Cell Carcinoma and Pseudoepitheliomatous Hyperplasia (Abu Eid and Landini, 2006)

The complexity of 57 ECTI profiles from nine SCC and 12 PEH-GCT cases were analyzed. The mean box fractal dimension of the ECTI profiles was 1.32 for GCT-PEH and 1.26 for SCC. When using large window sizes (190µm-557µm), the distribution of both local and local connected fractal dimensions showed that ECTI profiles were more complex in GCT-PEH in comparison to SCC profiles (Figure 7). However with the smaller window sizes (47µm -96µm) the local and local connected fractal dimension distributions were similar in both groups. Up to 82.5% (hierarchical discriminant analysis with leave-one-out) of the GCT-PEH and SCC profiles were correctly classified using the fractal descriptors (global and local).

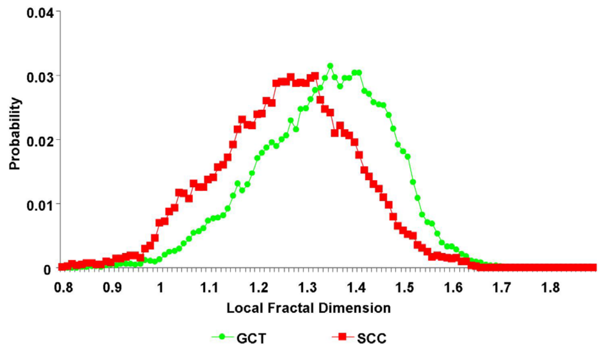


Fig. 7. Local fractal dimension probability of the ECTI for GCT-PEH and SCC at window size 179 pixels (557µm).

The results showed that ECTI profiles of GCT-PEH cases were significantly more complex than those of SCC. This may be due to active epithelial growth towards the underlying stroma and a concurrent growth by the granular cells towards the epithelium. In GCT with PEH, cell mixing at the ECTI may induce a different type of mechanical dynamics than those observed in epithelial SCC producing a more irregular ECTI.

3.6 Ageing of the Normal Oral Mucosa (Abu Eid et al., 2008)

Forty two ECTI profiles from 42 normal oral mucosa samples representing 10 different age groups (each group represented a decade in life ranging from the first to the ninth decade) were analyzed. The mean box fractal dimension for different age groups showed no significant differences (Figure 8).

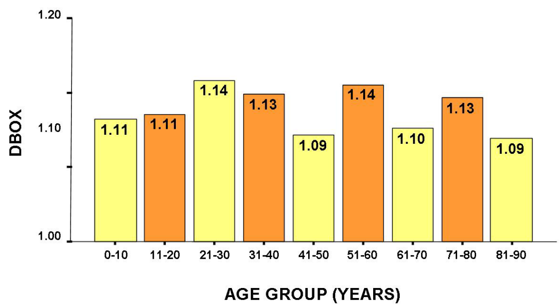


Fig. 8. Mean box fractal dimension (DBOX) of different age groups.

Various age-related changes in the epithelium have been described qualitatively in the literature, including the rete-ridges becoming less prominent and the ECTI profiles becoming smoother (Squier et al., 1976, Yemm et al., 1994), but preliminary quantitative results obtained in this study indicate that ageing of the oral mucosa does not seem to affect significantly the complexity of the epithelial connective tissue interface (at least in the range of scales investigated).

4. CONCLUSIONS AND FURTHER WORK

From this review, it is clear that different measures of fractal geometry are useful in the study of physiological and pathological changes in the oral mucosa. The tissue architectural changes manifested in the variations in the irregularity of the ECTI profiles can provide useful information for understanding pathological changes.

The methodologies used in this review can be expanded to study other pathological entities both inside and outside the oral cavity. Combined with cellular morphometric analysis and molecular analysis, further understanding of pathological processes can be achieved.

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