Laser Induced Fluorescence Spectroscopy of Soft Tissues of the Oral Cavity

Ajeetkumar Patil¹, Unnikrishnan V.K.¹, Rodney Bernard¹. Keerthilatha M.Pai², Ravikiran Ongole³, V.B.Kartha¹ and Santhosh Chidangil¹

¹Centre for Atomic and Molecular Physics, Manipal University, Manipal; ²Manipal College of Dental Sciences, Kasturba Medical College, Manipal University, Manipal; ³Manipal College of Dental Sciences, Kasturba Medical College, Manipal University, Mangalore, KARNATAKA, India-576104. santhosh.cls@manipal.edu

Abstract. The present study deals with the in vivo measurement of auto-fluorescence from different anatomical sites of oral cavities of healthy volunteers, using a homebuilt Laser Induced Fluorescence (LIF) Spectroscopy setup. Excitation wave length of 325 nm from a He-Cd laser was used as the source. From the 7 anatomical sites (say buccal mucosa, tongue, palate etc) of each oral cavity of 113 subjects, 1266 fluorescence spectra were recorded. The spectra were analysed using Principal Component Analysis (PCA) to see the correlation between different sites.

Keywords: Fluorescence, Oral cavity, PCA.

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INTRODUCTION

Oral cavity is one of the useful openings in our body, which describes a broad array of parts within the mouth including the lips, lining on the lips and cheeks referred to as buccal mucosa, teeth, tongue, floor of the mouth under the tongue, palate and the gums. With a complex structure, it performs many important nutritional, communicative and respiratory functions. It also known as the first portion of digestive tract where mechanical digestion will start by breaking food into small by mastication process. Oral cavity is much exposed to the outer environment than any other organ of the body. Due to this, oral cavity will greatly affect the routine function of our body followed by some diseases such as fibrillations, ulcers, cancer etc. Cancer is one of the intense among all diseases and considered to be a major threat to the public health [1]. It is the most common cancer in India accounting for 50-70% of total cancer mortality [2]. The survival rates are poor and not improved in last decade.

It is very well recognized that early detection is essential for successful therapy in all types of malignancy. Since, taking biopsies in the early stages of cancer may not be reliable [3] there is a need of some objective screening which will fill this gap to improve the survival rates. As there are many fluorophores are present in tissues, for eg, Pyridoxine, Collagen, Elastin, NADH, Porphyrins, and Flavins etc which have strong absorption bands in the 300-500nm range, giving rise to fluorescence in the 350-700nm range. In the present study, a homebuilt LIF set up was used to study different anatomical sites of oral cavity from 113 normal subjects. It will help in understanding the biochemical variations in tissue due to disease condition such as cancer.

MATERIALS AND METHODS

Figure 1: Block diagram of LIF Set-up

Figure 1 shows the block diagram of LIF prototype system. LIF system includes He-Cd Laser (325 nm), optical components, optical fibers, calibration source, SS fiber optic probe for oral cavity, spectrograph and CCD. 325nm laser beam was used to excite the tissue,
which is coupled to a single optical fiber which will carry laser light to the central fiber of the optic probe. The fiber optic probe is having a seven fiber coaxial arrangement, where central fiber for the excitation of the oral tissue and surrounding 6 fibers will collect the fluorescence emitted by the tissue which is fed to a spectrograph with CCD and then recorded in the computer.

**RESULTS AND DISCUSSION**

Typical fluorescence recorded from anatomical sites of oral cavity are shown in figure 2. The principal component analysis (PCA) of fluorescence spectra was done using PLS PLUS/IQ software from Thermo Galactic Corporation. PCA of 1269 fluorescence spectra of different anatomical sites (classes) are discussed below. PCA suggested that 12 factors to explain the spectra. But 5 factors are more than enough to simulate any LIF spectrum.

![Figure 2: Average spectra of Different anatomical sites of the oral cavity](image)

![Figure 3: Distribution of scores of Factor 1 for different anatomical sites](image)

Figure 3 shows the score distributions for different anatomical sites. From the figure it is clear that buccal mucosa, lip and tongue bottom shows positive scores, whereas palate, tongue tip and tongue top have negative scores with slightly less negative score for tongue lateral. To get a more accurate evaluation, Match/No Match test was performed using three parameters-scores, spectral residual, and Mahalanobis Distance.

<table>
<thead>
<tr>
<th>Sl.No.</th>
<th>Anatomical Site</th>
<th>Total No. of Spectra</th>
<th>No. of Matching Spectra</th>
<th>Resemblance (in %)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Buccal Mucosa</td>
<td>453</td>
<td>429</td>
<td>94.7</td>
</tr>
<tr>
<td>2</td>
<td>Tongue lateral</td>
<td>226</td>
<td>208</td>
<td>92.0</td>
</tr>
<tr>
<td>3</td>
<td>LIP</td>
<td>183</td>
<td>176</td>
<td>96.2</td>
</tr>
<tr>
<td>4</td>
<td>Tongue Tip</td>
<td>113</td>
<td>104</td>
<td>92.0</td>
</tr>
<tr>
<td>5</td>
<td>Tongue Top</td>
<td>118</td>
<td>35</td>
<td>29.7</td>
</tr>
<tr>
<td>6</td>
<td>Tongue Bottom</td>
<td>112</td>
<td>110</td>
<td>98.2</td>
</tr>
<tr>
<td>7</td>
<td>Palate</td>
<td>64</td>
<td>16</td>
<td>25.0</td>
</tr>
</tbody>
</table>

Standard set for Buccal mucosa was formed and tested against all the samples. Test results are given in Table 1. The test results showed that, there is a strong resemblance with buccal mucosa and all other classes except for two (i.e. palate and tongue top). This differentiation is expected due to the morphological and biochemical variations present in palate and tongue top. If we consider anatomy of these two classes, tongue top is covered by a specialized epithelium (mosaic of keratinized and nonkeratinized epithelium), whereas, soft palate composed of mucous membrane, muscular fibers and mucous glands.

**CONCLUSION**

The results presented here shows that Laser Induced Fluorescence is a powerful tool to differentiate between anatomical sites of oral cavity. Similar data for pathologically certified pre-malignant and malignant anatomical sites will lead to the development of highly objective screening tool for the early detection of oral cancer.

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**REFERENCES**