Evaluation of OSMF with Ultrasonography

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Abstract
Aims: To measure the submucosal thickness in Oral Submucous Fibrosis patients and in normal healthy individuals and to evaluate the changes in the vascularity by using color Doppler in different stages of Oral submucous fibrosis and in normal healthy individuals.

Materials and Method: The study group included a total of 50 male subjects out of these 40 OSMF were clinically diagnosed as having OSMF and they are further subdivided into 4 subgroups on the basis of clinical grading (according to Khanna and Andrade)each having 10 patients. While 10 apparently healthy subjects were included as controls. Submucosal thickness of labial and buccal mucosa was measured ultrasonographically for both the groups. Statistical analysis was done using mean ±SD, one way ANOVA test to compare the variable data.

Results: When the cases were compared with the controls there was significant increase seen in all the measured sites in cases. The increase in the thickness was found to be increase with the severity of the disease.

Conclusion: Significant positive relation was observed in interincisal opening with the duration of habit. However no correlation was observed with respect to frequency of habit and interincisal opening. There was increase in submucosal thickness as the severity of disease increased. There was decrease in vascularity in OSMF as compare to controls. Partial or complete absence of vascularity was seen in Grade III and Grade IV OSMF. A slight thickening of submucosa and decrease in vascularity may be considered as early changes seen in OSMF.

Keywords: OSMF, USG, Submucosal thickness, Vascularity

Introduction
OSMF is one of the most common premalignant condition in India, it is easy to diagnose but difficult to manage. Presently it is considered as an incurable and irreversible disease that progress even after cessation of habit.(1)

OSMF has been reported almost exclusively among Indians living in India and among other Asians with a reported prevalence ranging up to 0.4% in Indian rural population.

This condition affects approximately 0.5% (5 million people) of the population in the Indian subcontinent. The major presenting complaint is a progressive inability to open the mouth due to the accumulation of inelastic fibrous tissue in the juxta-epithelial region of the oral mucosa. This severely impairs eating and oral hygiene care. The epithelium overlying the fibrous condensation becomes atrophic in 90% of cases and is the site of malignant transformation in 4.5% of patients.(2)

It is an insidious disease primarily affecting buccal mucosa, soft palate and floor of the mouth. Burning sensation on eating spicy foods, superficial mucoceles (vesicles) and pale mucosa are some of the early signs. Bands of fibrosis appear which gradually widen to form sheets of fibrosis producing trismus which is progressive and irreversible.(3)

Evidence suggests that chewing of areca nut, pan masala and gutkha has been implicated in the development of OSMF. Areca nut extract could act as a potent stimulator for collagen synthesis in human fibroblasts culture leading to excessive accumulation of collagen leading to fibrosis and tannins present in areca nut reduced the degradation of collagen by collagenase, thus leading to OSMF.(4)

Since the habit of betel quid chewing and other commercially available products are wide spread in our country we need techniques that can be helpful for mass screening of this condition at an early stage. Ultrasonography is easy to use for the detection of soft tissue related diseases in oral and maxillofacial region. The ultrasound image indicates the surface structure of the lesions. Recently, Color Doppler ultrasound images using the Doppler effect of flow in blood vessels have also been applied to evaluate the presence or absence of vascular flow in normal tissues and in the diseased tissue.(5)

The USG system permits the evaluation of differences in the echogenicity of mucosal structures at a depth of up to 7 mm (24-fold magnification). This means that the zones of diagnostic interest are covered, i.e. epidermis, corium, and subcutaneous fatty tissue.(6)

In OSMF USG demonstrates the number, length, thickness of the fibrotic bands and pattern of overall vascularity in the affected area. The USG scan constitutes a full representation of the cross section of the buccal and labial mucosa, in the submucosal and muscular planes. The mucosal lining appears as a hyperechoic line, and the submucosa as a hypoechoic band supported by muscles planes. The band of
hypoechogenicity between the hyperechoic mucosa and the muscle layer is measured as submucosa.\(^{(7,8)}\)

This study aims to utilize the USG modality to detect the changes in OSMF which cannot be diagnosed clinically and to assess the submucosal thickness and vascularity of the oral mucosa in different stages of the disease.

**Materials and Method**

It is a case-control study in which 40 clinically diagnosed patients of OSMF is selected as cases and they were further subdivided into four groups 10 cases each on the basis of their clinical staging and 10 healthy individuals without any mucosal findings as controls.

Source of data is obtained from the Department of Oral Medicine And Radiology, Chhattisgarh Dental College and Research Institute, Sundara, Rajnandgaon.

**Inclusion Criteria**

**For Cases**
- Patients with positive history of arecanut or its product chewing.
- Clinical features with burning sensation on eating spicy foods, difficulty in swallowing and chewing, blanching of oral mucosa, palpable fibrous bands, restricted mouth opening.
- Age <20yrs>40yrs.

**For Controls**
- Normal healthy individual without any history of arecanut chewing.

**Exclusion Criteria**
- Patients with chronic illness, hypertension, pregnancy, and any kind of allergy.

A detailed history was obtained from the participants before undergoing USG examination. Clinical examination was done in dental chair with proper illumination, shown in Fig. 1. Clinical grading was given on the basis of interincisal mouth opening according to classification system given by Khanna and Andrade 1995.\(^{(9)}\)

Ultrasonographic imaging was performed by general radiologist with patient in supine position. The device used was ALOKA (Hitachi Aloka Medical Ltd.) with transducer probe of 3-12Mhz frequency. The probe was placed softly on the area so that it should not compress the tissues while performing USG and to obtain accurate measurements of the mucosa, shown in Fig. 2.

The submucosal thickness of both right and left buccal mucosa and upper and lower labial mucosa was performed. The mucosal lining was represented as hyperechoic line and the submucosa as hypoechoic band between the mucosa and the muscle plane, shown in Fig. 3. Vascularity was accessed by using color Doppler as shown in Fig. 4. This procedure is done in both cases and control groups.

**Statistical Analysis:** All measurements were taken in centimeters and echogenicity pattern was noted. The mean measurement was taken for right and left buccal mucosa. The data was subjected to statistical analysis using one way ANOVA test.

**Duration of habit:** In study Group 11(27.5%) out of 40 subjects were having habit since <5 years, 19(47.5%) patients were chewing since 5-10 years, 5 (12.5%) were chewing since 10-15 years. 5 (12.5%) patients were chewing more than 15 years.

**Frequency of habit:** Considering the frequency of habit 10 (25%) patients chewed between 1-5 times per day, 12 (30%) patients chewed between 5-10 times per day, 9 (22.5%) patients chewed between 10-15 times per day and 9 (22.5%) patients chewed more than 15 times per day.
Symptoms of the disease: In present study 10 (25%) of cases complained of burning sensation on having spicy food. 25 (67.5) cases complained of burning sensation with restricted mouth opening. 3 (7.5%) patients complained of restricted mouth opening and 2 (5%) patients complained of restricted mouth opening with difficulty in deglutation. 

Intraoral Examination: On clinical examination maximum interincisal opening ranged from 10-43 mm with mean of 28.92 mm. Blanching was present in all 40(100%) cases. Blanching with unilateral fibrous band were present in 8(20%) cases. Labial bands present in 9(22.5%) cases, tongue involvement in 4 (10%) cases, floor of the mouth and soft palate involvement were seen in 4 (10%) cases. Considering the duration of habit in study group of 40 patients, 11(27.5%) chewed for <5 years with the range of interincisal opening of 20-43 mm and mean 33.36 ± 7.59. 19(47.5%) patients chewed for 5-10 years, with the range of interincisal opening between 15-44 mm and mean of 30.42 ± 9.39. 5(12.5%) patients chewed for 10-15years with the range of interincisal opening between 10-43 mm and mean of 22.40 ± 12.85. 5(12.5%) patients chewed for more than 15 years with the range of interincisal opening between 11-36 mm and mean of 20 ± 9.56.

Significant difference was observed in mean values of Interincisal opening on the basis of duration of habit. The reported F=3.27 confirms this finding as it is statistically significant at 0.5 level (F=3.27, p<.05).

Tabulated value of F df (3,36) at 0.05 is 2.87, at 0.05 level 4.38 shown in Graph 1.

Table 1: Distribution of Age among Study Groups

<table>
<thead>
<tr>
<th>Age (years) ± SD</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
<th>Group 3</th>
<th>Group 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean</td>
<td>26.4±5.0</td>
<td>28.4±5.6</td>
<td>26.1±4.9</td>
<td>28.4±4.0</td>
<td>25.9±5.6</td>
</tr>
<tr>
<td>Range</td>
<td>19-34</td>
<td>20-40</td>
<td>23-43</td>
<td>21-30</td>
<td>18-40</td>
</tr>
</tbody>
</table>

Patients in Group 1 were within the age range of 19-34 years with a mean age of 26.4 ± 5.08. The maximum number of patients 6(60%) were in age group 20-25. In Group 2 all patients were male with the age range of 20-40 years with the mean age of 28.4 ± 5.60. The maximum number of patients 5(50%) were in age group between 25-30. In Group 3 age range of patients were 23-43 years with mean age of 26.1 ± 9.85. In Group 4 and 5 age range were 21-30 and 18-40.

Table 2: Correlation of Duration of habit with interincisal opening

<table>
<thead>
<tr>
<th>Duration</th>
<th>N</th>
<th>Mean</th>
<th>S.D.</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>&lt;5 yrs</td>
<td>11</td>
<td>33.36</td>
<td>7.59</td>
<td>20.00-43.00</td>
</tr>
<tr>
<td>5-10 yrs</td>
<td>19</td>
<td>30.42</td>
<td>9.39</td>
<td>15.00-44.00</td>
</tr>
<tr>
<td>10-15 yrs</td>
<td>05</td>
<td>22.40</td>
<td>12.85</td>
<td>10.00-43.00</td>
</tr>
<tr>
<td>More than 15 yrs</td>
<td>05</td>
<td>20.00</td>
<td>9.56</td>
<td>11.00-36.00</td>
</tr>
</tbody>
</table>

ANOVA Summary

Correlation of frequency of habit and interincisal opening was done. Frequency of habit was divided into 4 groups, 1-5 times, 5-10 times, 10-15 times, >15 times. 10(25%) cases chewed 1-5 times per day and there average interincisal opening ranges between 15-44mm and mean interincisal opening of 30.50 mm. 12(30%)cases chewed for 5-10 times a day with the range of interincisal opening of 20-41mm with mean of 31.58mm. 9(22.5%) cases had frequency of chewing between 10-15 times per day and their average interincisal opening ranged between 10-42 mm with mean of 10.64 mm. 9 (22.5%) cases had frequency of chewing more than 15 times per day with average interincisal opening of 11-40 mm and mean of 25.66 mm.

No statistically significant difference was observed in interincisal opening scores on the basis of frequency of habit. F=0.76, p=0.523.

Table 3: Correlation of frequency of habit with interincisal opening

<table>
<thead>
<tr>
<th>Frequency</th>
<th>N</th>
<th>Mean</th>
<th>S.D.</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>1-5 times</td>
<td>10</td>
<td>30.50</td>
<td>12.79</td>
<td>15.00-44.00</td>
</tr>
<tr>
<td>5-10 times</td>
<td>12</td>
<td>31.58</td>
<td>7.19</td>
<td>20.00-41.00</td>
</tr>
<tr>
<td>10-15 times</td>
<td>19</td>
<td>26.88</td>
<td>10.64</td>
<td>10.00-42.00</td>
</tr>
<tr>
<td>More than 15 times</td>
<td>10</td>
<td>25.66</td>
<td>10.44</td>
<td>11.00-40.00</td>
</tr>
</tbody>
</table>

ANOVA Summary

Comparison of Int. opening among Various Study Groups on the basis of frequency of habit

ANOVA Summary

Comparison of Interincisal opening among Various Study Groups on the basis of duration of habits

<table>
<thead>
<tr>
<th>Source</th>
<th>Df</th>
<th>Sum of squares</th>
<th>Mean square</th>
<th>F</th>
<th>Sig.</th>
</tr>
</thead>
<tbody>
<tr>
<td>Between Groups</td>
<td>03</td>
<td>870.398</td>
<td>290.13</td>
<td>3.27</td>
<td>.03</td>
</tr>
<tr>
<td>Within Groups</td>
<td>36</td>
<td>3192.377</td>
<td>88.677</td>
<td>2</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>39</td>
<td>4062.775</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Correlation of frequency of habit and interincisal opening was done. Frequency of habit was divided into 4 groups, 1-5 times, 5-10 times, 10-15 times, >15 times. 10(25%) cases chewed 1-5 times per day and there average interincisal opening ranges between 15-44 mm and mean interincisal opening of 30.50 mm. 12(30%)cases chewed for 5-10 times a day with the range of interincisal opening of 20-41 mm with mean of 31.58 mm. 9(22.5%) cases had frequency of chewing between 10-15 times per day and their average interincisal opening ranged between 10-42 mm with mean of 10.64 mm. 9 (22.5%) cases had frequency of chewing more than 15 times per day with average interincisal opening of 11-40 mm and mean of 25.66 mm.

No statistically significant difference was observed in interincisal opening scores on the basis of frequency of habit. F=0.76, p=0.523.
Though a slight decrease is seen as the frequency increases (>10times).

### Table 4: Showing Comparison of ultrasonographic measurements (cms) of submucosal thickness in various study groups & control group

<table>
<thead>
<tr>
<th>Group</th>
<th>No of Cases</th>
<th>ULM</th>
<th>LLM</th>
<th>RBM</th>
<th>LBM</th>
</tr>
</thead>
<tbody>
<tr>
<td>Group 1</td>
<td>10</td>
<td>0.17±0.02</td>
<td>0.18±0.02</td>
<td>0.20±0.04</td>
<td>0.20±0.03</td>
</tr>
<tr>
<td>Group 2</td>
<td>10</td>
<td>0.27±0.03</td>
<td>0.30±0.05</td>
<td>0.30±0.07</td>
<td>0.32±0.09</td>
</tr>
<tr>
<td>Group 3</td>
<td>10</td>
<td>0.40±0.05</td>
<td>0.41±0.08</td>
<td>0.46±0.07</td>
<td>0.54±0.13</td>
</tr>
<tr>
<td>Group 4</td>
<td>10</td>
<td>0.39±0.09</td>
<td>0.44±0.10</td>
<td>0.61±0.15</td>
<td>0.65±0.21</td>
</tr>
<tr>
<td>Group 5</td>
<td>10</td>
<td>0.17±0.01</td>
<td>0.16±0.02</td>
<td>0.19±0.02</td>
<td>0.18±0.02</td>
</tr>
<tr>
<td>F</td>
<td></td>
<td>46.09</td>
<td>34.84</td>
<td>41.88</td>
<td>28.75</td>
</tr>
<tr>
<td>p-value</td>
<td></td>
<td>.000</td>
<td>.000</td>
<td>.000</td>
<td>.000</td>
</tr>
</tbody>
</table>

USG measurement of submucosal thickness was done in 40 subjects on both right and left buccal mucosa and upper and lower labial mucosa. The mean of submucosal thickness for ULM was 0.17±0.02cm in Group 1 and 0.27±0.03cm in Group 2, in Group 3 it was 0.40±0.05cm and 0.39±0.09cm in Group 4. The mean submucosal thickness for LLM was 0.18±0.02 cm in Group 1, for Group 2 it was 0.30±0.05 cm, for Group 3 it was 0.41±0.08cm and for Group 4 it was 0.44±0.10cm. The mean submucosal thickness for RBM was Group 1, Group 2, Group 3 and Group 4 were 0.20±0.04 cm, 0.30±0.07 cm, 0.46±0.07cm, 0.61±0.15 cm respectively. Increase in the submucosal thickness was observed with the severity of the disease. Significant difference was observed on mean values of ULM, LLM, RBM and LBM among study groups. The reported F and p value confirms the findings as statically significant.

### Table 5: Comparison of Vascularity seen in color Doppler

<table>
<thead>
<tr>
<th>Vascularity</th>
<th>N</th>
<th>Group 1</th>
<th>Group 2</th>
<th>Group 3</th>
<th>Group 4</th>
<th>Group 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>Seen</td>
<td>10</td>
<td>(40%)</td>
<td>(30%)</td>
<td>(10%)</td>
<td>(20%)</td>
<td>(80%)</td>
</tr>
<tr>
<td>Partially Seen</td>
<td>10</td>
<td>(50%)</td>
<td>(40%)</td>
<td>(50%)</td>
<td>(30%)</td>
<td>(20%)</td>
</tr>
<tr>
<td>Absent</td>
<td>10</td>
<td>(10%)</td>
<td>(30%)</td>
<td>(40%)</td>
<td>(50%)</td>
<td>0</td>
</tr>
</tbody>
</table>

In Group 1 Vascularity is seen in (40%) cases, (50%) cases showed partial vascularity and in (10%) case in was absent. In Group 2 vascularity in seen in (30%) cases, (40%) cases showed partial vascularity while in 3 (30%) cases it was absent. In Group 3, (10%) showed vascularity, (50%) cases showed partial vascularity while in (40%) cases it was absent. In Group 4 vascularity is seen in (20%) cases and in (30%) it was partially seen and in (50%) cases it was absent. In Group 5 vascularity was present in all subjects.

### Discussion

OSMF is one of the most prevalent premalignant condition that occurs predominantly among Indians and people of Indian origin living outside India, occasionally in other Asians, and sporadically in Europeans. In India, OSMF has a wide range of prevalence rate of 0.2% to 1.2% and a malignant transformation rate of 3-7.6%.(3)

Fibrosis in OSMF is a dynamic process and needs periodic monitoring of disease activity, to assess response to cessation of habit or other medical interventions.(7)

Biopsy, being minor surgical procedure is associated with some discomfort to the patient and is time consuming. Besides biopsy does have its limitation in a diffuse disease like OSMF, where a tissue sample obtained from single site may not be representative of the true extent of the severity of disease. Also there are chances that there is scarring and increased fibrosis at the actual site of biopsy.(10)

Ultrasoundography is particularly suitable for imaging superficial structures of head and neck region. It provides both qualitative and quantitative assessment. It has the ability to detect the nature of lesion, its dimensions, distance from the skin surface and relative proximity to mucosal structures.(7)

**Duration of Habit:** Considering the duration of habit in the present study Table 2 shows that significant decrease in the interincisal opening as the duration of habit increases. Eipe et al.(2005) study reported that habitual use of betel quid for >5 years predisposes the oral mucosa to oral premalignant disorders including OSMF.(10)

Maher et al. also reported that individuals using smokeless tobacco products with arecanut for up to a decade are more susceptible to develop OSMF compared with subjects using such products for a shorter duration. (11) Similarly, a recent case-control study reported that gutka-chewing habit for up to 4 years increases the relative risk of developing OSMF. (12) As there is constant contact between the
mixture of arecanut and oral mucosa which causes repeated microtrauma by the friction of coarse fibres of arecanut which facilitates diffusion of the alkaloids into subepithelial connective tissue resulting in juxtaepithelial inflammatory cell infiltration.(13)

In contrast to the present study, Poornima R (2010) found no correlation between duration of chewing habit and clinical or histological grade of OSMF. They suggested that daily consumption was more significant than total duration of habit and daily consumption rate appears to be much more significant with respect to risk than lifetime duration of the habit.(9)

Frequency of habit: While correlating the frequency of the habit to interincisal opening, Table 2 shows no significant association between the frequency of habit and interincisal opening. This is compatible with few studies conducted by Gupta DS et al(1980), Canniff JP et al(1986), Kuttan R et al (1981) they stated that duration was more significant than the frequency of the habit.(14)

Correlation of Submucosal Thickness in OSMF cases and in controls: The most important parameters describing the interactions between ultra- sound and the tissue through which it is transmitted are attenuation, velocity and impedance. The attenuation and velocity increase in proportion to the relative amounts of collagen in the tissue. On the other hand, they decrease in relation to the increase in water content. Collagen has a greater modulus of elasticity than other tissues. This leads to a higher velocity and higher impedance and for this reason collagen is one of the main sources of echogenicity. As there is increase in the collagen fibres in OSMF this could be the most possible reason of increase echogenicity and increased submucosal thickness in USG.(8)

The current study showed an increase in submucosal thickness from Grade I to Grade IV in ULM, LLM, RBM, LBM (Table 4). Similar result was found in another study by Poornima R et al in 2010.(8)

In this study the mean thickness of ULM in cases is found to be less than LLM, in Group IV which is the most advanced OSMF group the mean thickness of ULM was 0.39cm and in LLM it was 0.44 cm. The possible reason behind this could be that most patients have habit of placement of arecanut in the lower labial vestibule, which increases the collagen activity of that particular site more than that of other sites in the oral cavity. The reason for increase in the submucosal thickness could be due to increase collagen activity and increase activity of proliferation of fibroblast in the disease process.

Correlation of Vascularity seen in Color Doppler: Any changes in vascularity seen in color Doppler in OSMF patients and controls were noted according to the grades of OSMF and controls.

In Grade I, vascularity is seen in 4(40%) cases, 5(50%) cases showed partial vascularity and in 1(10%) case in was absent. In Grade II vascularity in seen in 3(30%) cases, 4(40%) cases showed partial vascularity while in 3 (30%) cases it was absent. In Grade III, 1(10%) showed vascularity, 5(50%) cases showed partial vascularity while in 4(40%) cases it was absent. In Grade IV vascularity is seen in 2(20%) cases and in 3(30%) it was partially seen and in 5(50%) cases it was absent. Vascularity was seen in all subjects of control group.

A slight decrease in vascularity was noted in Grade I OSMF ass compare to control. This may be considered as the initial change in Grade I OSMF when compared to controls.

Decrease in the vascularity was seen in color Doppler USG with the progression the disease. This supports the findings of another study conducted by Manjunath et al.(6)

Conclusion

As USG is a non-invasive technique which the patient accepts readily, and comparatively a cheaper diagnostic modality.

Studies are advised to evaluate the applications of USG to detect the submucosal thickness and to compare it with healthy subjects and OSMF patients. In our study we had taken only submucosal area which has been replaced by fibrosis. But in OSMF there is involvement of underlying muscles too, which may be one of the factor for trismus hence studies are advised to measure the submucosal thickness along with muscle layer. For precise evaluation application of intraoral probe is advice.

References