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## **Original Research**

# Estimation of hemoglobin, serum iron, total iron-binding capacity and serum ferritin levels in oral submucous fibrosis

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#### ABSTRACT:

**Background**: The present study was aimed to evaluate Hb, serum iron, total iron-binding capacity (TIBC) and serum ferritin in OSMF patients. **Material and methods**: Hb, serum iron, total iron binding capacity (TIBC), and serum ferritin levels were measured in 50 OSMF cases and compared to those of healthy controls. T-tests, one-way analyses of variance, and Tukey's multiple post hoc tests were used to compare the outcomes. **Results**: Out of 50 OSMF patients, according to clinical staging, Stage-I were 15 cases (30%), Stage-II were 25 cases (50%) and Stage-III were 10 cases (20%), and according to histopathological grading, early stage were 15 cases (30%), intermediate stage were 25 cases (50%) and advanced stage were 10 cases (20%). A control group of forty normal sex- and age-matched subjects were also included in the study. Hb, serum iron, TIBC and serum ferritin values were evaluated in both study group and the control group. **Conclusion**: In OSMF patients, relative to controls and with increasing clinical stage and histological grade of OSMF, Hb, serum iron, and serum ferritin levels were lowered, but TIBC increased.

Keywords: serum iron, oral submucous fibrosis

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#### **INTRODUCTION**

Oral submucous fibrosis (OSMF) is a chronic and latent malignant disease, which poses a global and regional problem to public health, especially in East and Southeast Asia where areca nut chewing is popular. The malignant transformation rate of OSMF to oral squamous cell carcinoma (OSCC) accounts for 7%-13% [1].

Fibrosis and hyalinization of subepithelial tissue are the most crucial clinicopathological features of OSMF, which greatly affect the patients' quality of life. The appearance of myofibroblasts and the consistent expression of  $\alpha$ -smooth muscle actin ( $\alpha$ -SMA) are considered to be signs of progressive fibrosis and are thought to cause the change of OSMF microenvironment, leading to tumorigenesis [2].

Even though the exact etiology and pathogenesis of OSMF have not yet been elucidated totally, the currently recognized pathogenic factor is areca nut. Meanwhile, great progress of OSMF-relevant diagnosis and clinical treatment has been reported in recent years.OSMF is also considered as an Asian version of sideropenic dysphagia, wherein chronic iron deficiency leads to mucosal susceptibility to irritants, such as chillies and areca nut products. Hemoglobin (Hb) levels, in particular, serum iron levels, are considered as biochemical indicators for nutritional assessment.[3]

Hence, the present study was aimed to evaluate Hb, serum iron, total iron-binding capacity (TIBC) and serum ferritin in OSMF patients.

#### MATERIAL AND METHODS

Objectives Hemoglobin (Hb), Serum Iron, Total Iron-Binding Capacity (TIBC), and Serum Ferritin were analyzed in this study on OSMF patients.Fifty individuals with OSMF and fifty healthy individuals served as controls in the current investigation. Patients' histories and clinical results were collected after receiving their informed consent. Both the clinical and histological findings were used to categorize these patients into stages (Stage-I: Mouth opening >45 mm; Stage-II: Restricted mouth opening 20-44 mm; Stage-III: Mouth opening 20 mm).Patients were recalled after histological confirmation, and 5 ml of venous blood was collected; 1 ml was immediately transferred to а vacutainer containing ethylenediaminetetraacetic acid (anticoagulant) for Hb analysis via the cyano Hb method, and the remaining 4 ml were allowed to clot before centrifugation for 5 minutes at 3000 rpm separated the serum from the blood. Tulip iron and TIBC kit and Bios Microwell ELISA Diagnostic Systems kit were used to analyze serum iron, TIBC, and serum ferritin levels, with the results processed in a photocolorimeter.

#### RESULTS

Out of 50 OSMF patients, 18 (36%) were classified as Stage I, 22% as Stage II, and 10% as Stage III clinically; similarly, 18 (36%) were classified as Early Stage, 22% as Intermediate Stage, and 10% as Advanced Stage histopathologically. The research also included a control group of forty healthy people of the same age and gender. Both the study group and the control group had their hemoglobin (Hb), serum iron, transferrin (TIBC), and serum ferritin levels measured.

Results showed that the mean Hb% (14.56), serum iron (82.63), and serum ferritin (189.41) levels in OSMF patients were significantly lower compared to those in controls (P = 0.00001), while the mean TIBC (342.3) levels in OSMF patients were significantly higher compared to those in controls (P = 0.00001).

Parameters	OSMF Group Mean value	Control group Mean value	P value
Haemoglobin	14.56	16.39	P <0.05 (significant)
Serum iron	82.63	98.36	
TIBC	342.3	201.33	
Serum ferritin	189.41	212.98	

However, mean TIBC levels indicated a gradual increase as the clinical stage of OSMF progressed, in contrast to the mean Hb%, serum iron, and serum ferritin levels. Using a one-way ANOVA test, we find that this difference is significant (P 0.0001). Clinical staging comparisons between Stage I and Stage II, Stage I and Stage III, and Stage II and Stage III yielded statistically significant results (\*P 0.05) when analyzed using Tukey's multiple post hoc procedures.

As the histologic grade of OSMF increases, the mean Hb%, serum iron, and serum ferritin levels all drop, with the exception of the mean TIBC levels, which increase. Using a one-way ANOVA test, we find that this difference is significant (P 0.0001). Tukey's multiple post hoc procedures found statistically significant differences in histological grading between the early and intermediate stages, the early and advanced stages, and the intermediate and advanced stages, respectively (\*P 0.05).

#### DISCUSSION

The overall prevalence of OSMF in India is about 0.5% with a range of 0.2%–1.2% in different regions of the country.[4] Recent epidemiological data indicate that the number of cases of OSMF has risen rapidly in India due to an upsurge in the popularity of commercially prepared areca nut preparations and an increased uptake of this habit by young people.[5]

The etiology of OSMF is multifactorial but areca nut chewing is the main causative agent. An equally important second aspect which needs to be considered is the preconditioning of the oral mucosa by a prolonged, chronic deficiency of iron and/or Vitamin B-complex, anemia and a genetic predisposition to the disease.[6] In the study performed by Tadakamadlaet al.[7] the mean copper and iron level differed significantly between the OSMF patients and controls, with patients exhibiting higher copper and lower iron levels in contrast to controls who presented lower copper (P < 0.005) and higher iron levels (P < 0.01). The results also showed that there was decrease in serum iron concentration as clinical stage increased (P < 0.0001). In the study conducted by Shetty et al.[8] the serum and salivary iron levels decreased in OSMF patients with the progression of histopathological grading, but this was not significant. Serum and salivary levels

showed significant correlation among cases (r = 0.315

and P = 0.011), but not among controls. Karthik H et al [9] estimated the hemoglobin and serum iron levels among patients with oral submucous fibrosis and to compare the values with healthy subjects. In this hospital-based study 30 diagnosed patients of OSMF and 15 healthy individuals were included, and the values of hemoglobin and serum iron levels were estimated using Sahli's and Ferrene methods. Results. **OSMF** patients showed significantly lower levels of hemoglobin and serum iron when compared with the healthy subjects. The findings of the study emphasizes on the assessment of hemoglobin and serum iron for patients with oral submucous fibrosis. Also, iron therapy should be instituted concomitantly with the initial diagnosis which helps to cease the further progression of the condition.

#### CONCLUSION

In OSMF patients, relative to controls and with increasing clinical stage and histological grade of

OSMF, Hb, serum iron, and serum ferritin levels were lowered, but TIBC increased.

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