

# The Excretion of Lactoferrin in Thai Patients with Oral Lichen Planus

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**Abstract—Introduction:** The infection of *Candida* in the oral cavity of oral lichen planus (OLP) patient undergoing steroid treatment have been reported continually. Lactoferrin is one of the salivary antimicrobial protein. The recent study clearly showed that the significant decrease of lactoferrin correlated with *Candida* growth in the oral cavity. Although there have numerous studies of lactoferrin but the relation of lactoferrin level in Thai patients with OLP who are not undergoing steroid treatment has never been reported. **Objective:** The aim of this study was to investigate the excretion of lactoferrin in the saliva of OLP patients who were not undergoing steroid treatment compared with healthy volunteers. **Methods:** Fifteen OLP patients who were not undergoing steroid treatment and fifteen healthy volunteers were recruited in this study. Whole unstimulated saliva were collected from all participants. Salivary flow rate was calculated and the excretion of salivary lactoferrin was analyzed using ELISA. **Results:** There was no statistical difference in the salivary flow rate of OLP patients and healthy volunteers ( $P=0.455$ ). Furthermore, the excretion of salivary lactoferrin in patients with OLP compared with healthy volunteers was not significantly different ( $P=0.694$ ). **Conclusion:** The results in this study revealed that there was no difference in the salivary flow rate and the excretion of salivary lactoferrin in Thai patients with OLP who were not undergoing steroid treatment compared with healthy volunteers.

**Keywords**—Lactoferrin, oral lichen planus, salivary flow rate

## I. INTRODUCTION

Oral lichen planus (OLP) is a chronic inflammatory disease of the oral mucosa predominantly presents in middle-aged or elderly women [1]. Many studies showed that the progression of OLP involved cell-mediated immunological dysfunction [2, 3]. The symptom of OLP such as burning sensation or pain usually affects the patients' quality of life. At present, the number of drugs were suggested to use in treatment of OLP. The most common drug that widely use is topical steroid [4]. However, the

relation of oral candidiasis with topical steroid using in OLP patients were reported [5, 6, 7].

Saliva contains multiple kinds of antimicrobial proteins and peptides that inhibit fungi such as lactoferrin (Lf), immunoglobulin A (IgA) and defensin [8, 9, 10, 11]. Lactoferrin is an iron-binding glycoprotein in transferrin family. It is considered to be a key component of the innate host defense system that exhibits strong antimicrobial activity against fungi, virus and broad spectrum of bacteria [12]. The mechanism and biological role of lactoferrin have been studied widely and a recent report clearly showed that the significant decrease of lactoferrin correlated with the growth of *Candida* in the oral cavity [13].

At present, the studies of lactoferrin had been reported continually. The decrease of salivary lactoferrin was revealed in patients with oral candidiasis and oral dryness [13, 14]. Nevertheless, the relation of lactoferrin level in Thai patients with OLP has never been reported. Hence, the aim of this study was to investigate the excretion of lactoferrin in the saliva of OLP patients who were not undergoing steroid treatment compared with healthy volunteers.

## II. MATERIALS AND METHODS

### *Patients and healthy volunteers*

Fifteen patients comprised 14 females and 1 male whose age range from 24 to 62 years ( $48.07 \pm 9.66$  years; mean  $\pm$  SD) with clinical and histopathological of OLP were enrolled in the present study. Furthermore, fifteen healthy volunteers comprised 15 females whose age range from 42 to 72 years ( $54.2 \pm 9.63$  years; mean  $\pm$  SD) presented at the Dental Hospital, Faculty of Dentistry, Chulalongkorn University, Bangkok, Thailand were recruited in this study. All patients and volunteers had no signs of oral candidiasis at the investigating time. Furthermore, all participants had no history with any diseases or medical conditions predisposing them to oral candidiasis or the promotion of *Candida* carriage and no history of taking antifungal agents, using antiseptic mouthwashes, or smoking at less 1 month. The study was performed with the approval of the Ethics Committee of the Faculty of Dentistry, Chulalongkorn University, and written informed



consent was obtained from all subjects before the start of the study.

#### *Saliva collection*

Whole unstimulated saliva was collected from all participants after the meal for at least 2 hours, by ejecting into the sterilized tube until 2 ml were obtained or 10 minutes had elapsed [14]. After salivary flow rate calculation, the collected saliva was centrifuged with refrigerated centrifuge machine at 16,000 rpm for 20 minutes at 4°C and was stored at -80°C before used in experiments.

#### *Lactoferrin assays*

In brief, 100 µl saliva samples were used to determine salivary lactoferrin by enzyme-linked immunosorbent assays (ELISA) method (lactoferrin ELISA kit; MyBioSource Inc; USA) performed according to the manufacturer's manual. Subsequently, the level of salivary lactoferrin was detected by spectrophotometer at 450 nm immediately. To compare with the standard curve, data were calculated the concentration of salivary lactoferrin and eventually converted into the excretion of salivary lactoferrin.

#### *Statistical analysis*

Statistical analysis was carried out using Statistical Package for the Social Science (SPSS) version 12.0. Mann-Whitney U test was used to analyze the statistical difference of the level of salivary lactoferrin between OLP and healthy participants. A *P*-value of less than 0.05 was considered statistically significant.

### **III. RESULTS**

#### *Salivary flow rate*

The salivary excretion of patients with OLP and healthy volunteers was evaluated. The salivary flow rate of OLP patients and healthy volunteers were  $0.36 \pm 0.21$  and  $0.43 \pm 0.26$  ml/min, respectively. There was no statistical difference in salivary flow rate between OLP patients and healthy volunteers (*P*=0.455) (table 1).

#### *Lactoferrin concentration and excretion*

To investigate the concentration and excretion of lactoferrin, unstimulated whole saliva was collected and analyzed using ELISA. The concentration of lactoferrin in saliva of OLP patients and healthy volunteers were  $3.65 \pm 3.25$  and  $2.29 \pm 1.13$  ng/ml, respectively. There was no statistical difference in lactoferrin concentration

between OLP patients and healthy volunteers (*P*=0.206) (table 1). The excretion of salivary lactoferrin in OLP patients and healthy volunteers were  $1.21 \pm 0.91$  and  $0.99 \pm 0.71$  ng/min, respectively. However, the statistic showed no significant difference in salivary lactoferrin excretion between OLP patients and healthy volunteers (*P*=0.694) (table 1).

### **IV. DISCUSSION**

Lactoferrin is one of the salivary antimicrobial protein that suppress the growth of oral microorganism [12]. The decrease in excretion of salivary lactoferrin closely correlates with low salivary flow rate [14]. Therefore, reducing salivary flow rate is one of the crucial factors that plays an important role in the growth of oral opportunistic microorganisms.

The salivary flow rate was investigated in our study, the result revealed no significant difference between OLP patients and healthy volunteers and none of our subjects showed the sign of oral dryness.

The infection of *Candida* in the oral cavity of OLP patients undergoing topical steroid treatment have been reported continually [5, 6, 7]. To use steroid is well known as a predisposing factor promoting the growth of *Candida* [15]. Nevertheless, the etiology of *Candida* infection in the oral cavity involves many factors. The change in excretion of salivary lactoferrin was elucidated as one of the factors involving the overgrowth of *Candida* in the oral cavity [13]. The level of salivary lactoferrin was investigated in this study. However, the excretion of salivary lactoferrin in this study revealed no difference between the group of OLP patients who were not undergoing steroid treatment and the group of healthy volunteers.

Oral *Candida* infection usually promotes the development in symptom of OLP patient such as pain or burning sensation. At present, numerous studies of lactoferrin were reported and the biological role of salivary lactoferrin in inhibiting the overgrowth of oral opportunistic microorganism was elucidated. However, no study of salivary lactoferrin in Thai OLP patients has been reported. In our study, we showed that there was no change in the excretion of salivary lactoferrin in OLP patients who were not undergoing steroid therapy. To elucidation of the cause of *Candida* infection in the oral cavity of OLP patients treated with topical steroid requires further study.



Table 1 Salivary flow rate and salivary lactoferrin

Excretion and concentration	OLP patients Mean $\pm$ SD	Healthy volunteers Mean $\pm$ SD	P - value
Salivary flow rate (ml/min)	0.36 $\pm$ 0.21	0.43 $\pm$ 0.26	0.455
Lactoferrin concentration (ng/ml)	3.65 $\pm$ 3.25	2.29 $\pm$ 1.13	0.206
Lactoferrin flow rate (ng/min)	1.21 $\pm$ 0.91	0.99 $\pm$ 0.71	0.694

## V. CONCLUSIONS

The results in this study revealed that there was no difference in the salivary flow rate and the excretion of salivary lactoferrin in Thai patients with OLP who were not undergoing topical steroid treatment compared with healthy volunteers.

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## CONFLICT OF INTEREST

The authors declare that there have no conflict of interest.

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