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## Determining Relative Percentage Change as a New Metrics in Scaling and Root Planning Therapy Outcome in Patients with Chronic Periodontitis

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#### Authors' contributions

This work was carried out in collaboration among all authors. Author ASA designed the study, managed the literature searches, wrote the protocol, estimated the biochemical markers, obtained the data and wrote the first draft of the manuscript. Author RK managed the clinical evaluation of the patients and performed SRP therapy on them. Authors MGK and ANS supervised and guided the study. All authors read and approved the final manuscript.

#### Article Information

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**Original Research Article** 

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#### ABSTRACT

**Aims:** To quantify the relative percentage change in the patients with periodontitis after employing the scaling and root planning (SRP) therapy and determine the association in the improvements in clinical parameters of biochemical oxidative stress (OS) markers, total antioxidant capacity (TAOC), and malondialdehyde (MDA).

**Study Design:** In this cross sectional study, two groups of patients were clinically evaluated and their biochemical parameters were quantified and statistically compared.

**Place and Duration of Study:** Departments of Biochemistry and Periodontology, M. A. Rangoonwala Dental College, Pune and Department of Biochemistry, Grant Govt. Medical College, Mumbai. The study was carried out between May 2010 and July 2012.

**Methodology:** Individuals with generalized chronic periodontitis (n = 86; CAL  $\geq$  3mm American Academy of Periodontology 1999 criteria) were clinically (Gingival index, plaque index, probing depth, clinical attachment level) and biochemically (TAOC, MDA) evaluated. The SRP therapy was performed and a follow-up was done after 3 months. The mean values of clinical and biochemical study parameters and their relative percentage change were evaluated thoroughly.

**Results:** Individuals with chronic periodontitis showed improved clinical and biochemical oxidative stress (OS) markers. After the SRP therapy, significant improvement (p < 0.05) was found in both clinical and biochemical parameters from their corresponding pre-treatment values.

The relative percentage change in clinical parameters ranged between 14.5% to 38.7%, and those of TAOC and MDA were -10.3% and 34.71% respectively. Further, the relative % change in TAOC and MDA showed significant correlation (p < 0.05) to those of PI, PD, and CAL.

**Conclusion:** On conducting the SRP therapy, improvement in clinical parameters and biochemical OS parameters is noticed in the individuals with periodontitis. The relative percentage change can be used as a tool for therapy outcome assessment.

Keywords: Scaling and root planning; relative percentage change; determining a possible metrics; SRP a tool for treating periodontitis; assessing clinical and biochemical parameters.

#### 1. INTRODUCTION

Oxidative stress (OS) is known to accelerate the progression of growing number of human diseases, [1] including periodontitis [2]. OS is linked to the onset of periodontal tissue destruction and progression of chronic periodontitis [CP] [3,4], an inflammatory disorder of the periodontium [5]. The pathology of periodontitis shows that there is an important role of reactive oxygen species [ROS], antioxidant [AO] systems, and the products of OS [5].

Dental professionals and periodontologists employ scaling and root planning (SRP) as the first phase in the periodontal therapy for controlling CP [6]. This therapy has proved to be effective in oral clinical parameters such as probing depth [PD], clinical attachment level [CAL] [6], and healthy outcomes in the levels of biochemical OS markers including total antioxidant capacity and lipid peroxidation etc. [5,7]. CP and OS have a bi-directional relationship [8]. The CP affects the OS status in the individuals. By controlling the CP through the application of the SRP therapy resulted into beneficial outcomes in the OS markers [9,10].

Generally studies in the literature have considered the mean values of the study parameters for assessing the therapy outcomes. The present study aims to quantify the association of the extent of improvement (relative percentage change) in clinical parameters to those of the levels of TAOC and MDA in the individuals with CP.

## 2. MATERIALS AND METHODS

#### 2.1 Study Groups

**Group I (C):** Individuals with slight chronic periodontitis (CAL < 3mm); n = 120 (males = 65, females = 55), Mean age =  $38.51 \pm 5.55$  comprises the control group. They were apparently healthy with good oral and systemic health and had no records of any habits or illness that would require medication or hospitalization in the last six months. They were clinically evaluated only once and their blood sample was collected and analyzed only once.

**Group II (CP):** Individuals with severe chronic periodontitis (CAL  $\geq$  3 mm); n = 86 (males = 51, females = 35), Mean age = 40.9 ± 4.61 belonged to this group. The subjects in this group were otherwise healthy, with no history of major illness and consumption of antioxidants, antibiotics, antiinflammatory, or any other drugs. They had not received any periodontal therapy for at least 6 months prior to the inception of the study. Individuals having past illness and undergoing any treatment, or those who had been diagnosed with diabetes or were alcoholics were excluded from the study. This group was given the SRP therapy and a follow-up was done after 3 months.

#### 2.2 Clinical Measurements

The individuals in both the study groups were clinically evaluated for chronic periodontitis according to the criteria of the American Academy of Periodontology (1999) [11]. The periodontal status of all individuals was evaluated by the measurement of gingival index (GI) [12], plaque index (PI) [13], probing depth (PD), and clinical attachment level (CAL). The PD and CAL measurements were done as prescribed in [11,14]. All clinical measurements were evaluated in compliance with the standard of the University of North Carolina (UNC-15) probe (Hu-Friedy, Chicago).

#### 2.3 Sample Collection

A total of 4 ml venous blood was collected in disposable syringes from all the subjects following routinely employed precautionary measures. Of this, 2 ml blood was transferred to sodium heparin vacutainer and the obtained plasma was used for TAOC estimation. The remaining 2 ml blood was left at room temperature for 30 minutes and centrifuged at 3,000 rpm for 20 minutes to obtain serum, which was used for the analysis of MDA. All the biochemical markers were measured on calibrated semi-auto analyzer BIOTRON BTR-830 (Ranbaxy Laboratories, India). The blood samples were collected twice; once at baseline (C and CP group) and then after 3 months of the SRP therapy (CP group).

#### 2.4 Biochemical Studies

#### 2.4.1 Total Anti-Oxidant Capacity (TAOC)

TAOC was measured with the help of the ferric reducing ability of plasma (FRAP) assay. At low pH when ferric tripyridyltriazine [Fe<sup>3+</sup>TPTZ] complex is reduced to its ferrous form [Fe<sup>2+</sup>TPTZ], an intense blue color with an absorbance maximum at 593 nm develops [15].

#### 2.4.2 Malondialdehyde (MDA)

The proteins in the serum are precipitated by trichloroacetic acid (TCA) and the mixture is heated with thiobarbituric acid in 2 M sodium sulphate in a boiling water bath for 30 minutes. The resulting chromogen is extracted with n - butyl alcohol and absorbance of organic phase is determined at 530 nm [16].

#### 2.5 SRP Therapy

The participants in group CP received periodontal therapy, which included SRP and oral hygiene instructions. SRP was performed by qualified periodontologists using ultrasonic instrument (Electro Medical System, Switzerland) and manual Gracey curettes (Hu-Friedy, Avco). The instructions for oral hygiene included brushing of teeth with Bass technique [17] twice daily after meals. Post SRP therapy, a follow-up was done after 3 months.

#### 2.6 Relative % Change Calculation

Relative % change Table 3 is calculated using the formula;

[[Baseline value – Post Treatment value] ÷ Baseline value] × 100

Negative value indicates post treatment higher value and vice versa.

#### 2.7 Statistical Analysis

The statistical analysis to study the parameters was done using Statistical Package for Social Sciences (IBM-SPSS version 19) in Microsoft Windows. The values were expressed as mean  $\pm$  SD across the study groups. The independent sample *t* test was employed for the comparison of the significance of the difference of the mean values. The correlation of relative % change in clinical and biochemical parameters was assessed using two-tailed Pearson correlation test. *P* value < 0.05 is considered to be statistically significant.

#### 3. RESULTS AND DISCUSSION

Oxidative stress (OS) has gained high importance in assessing human health and diseases. It is associated with both the onset of periodontal tissue destruction and systemic inflammation [3] with increased ROS concentration leading to oxidative damage and impaired circulating oxidant: anti-oxidant balance [18,19]. SRP is the basic therapy amongst all the periodontal therapies and is routinely employed in dental practice. It has shown beneficial outcomes in clinical and biochemical parameters in individuals with CP [20,21,22].

The obtained values indicate significantly higher mean values (p < 0.001) of clinical parameters and higher level of MDA. These values compromised TAOC in the individuals belonging to CP than C group Table 1.

Furthermore, the results Table 2 showed a statistically significant difference (p < 0.001) in the post treatment mean values of clinical and

biochemical parameters compared to their corresponding baseline (pre-treatment) values in the individuals of CP group. Table 3 represents the mean relative % change in the clinical and biochemical parameters in CP group.

The relative percentage change of TAOC and MDA showed significant (p < 0.05) negative correlation to PI and PD. TAOC showed negative correlation to CAL and reached close to statistical significance (p = 0.067) Table 4.

Table 1. Comparison of baseline clinical parameters and biochemical markers between c and
cp group

Clinical parameters	Mean ± SD		P <sup>*</sup> value
	Group C	Group CP	
	[n = 120]	[n = 86]	
GI	0.67 ± 0.11	2.39 ± 0.48	<0.001
PI	0.43 ±0.42	2.27 ± 0.52	<0.001
PD [mm]	1.73 ± 0.30	5.44 ± 0.44	<0.001
CAL [mm]	1.83 ± 0.30	7.68 ± 0.88	<0.001
Biochemical Oxidative Stress Markers			
TAOC [µM/L]	913.42 ± 66.0	826.34 ± 76.81	<0.001
MDA [nM/ml]	2.02 ± 0.24	4.11 ± 0.38	<0.001
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Values for mean ± SD, \* p values are obtained using independent sample t test, p values < 0.05 is considered to be statistically significant GI = Gingival Index, P I= Plaque Index, PD = Probing Depth, CAL = Clinical Attachment Level, TAOC = Total Anti-Oxidant Capacity, MDA = Malondialdehyde

Table 2. Comparison of baseline and post-treatment values of clinical parameters and	d
biochemical markers in cp group (n = 120)	

Clinical Parameters	Mean ± SD		P <sup>*</sup> value
	Baseline	Post-Treatment	
GI	2.39 ± 0.48	1.47 ± 0.48	<0.001
PI	2.27 ± 0.52	1.39 ± 0.49	<0.001
PD [mm]	5.44 ± 0.44	4.65 ± 0.51	<0.001
CAL [mm]	7.68 ± 0.88	6.10 ± 1.02	<0.001
Biochemical Oxidative Stress Markers			
TAOC [µM/L]	826.34 ± 76.81	910.0 ± 68.01	<0.001
MDA [nM/ml]	4.11 ± 0.38	2.70 ± 0.45	<0.001
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Values for mean ± SD, \* p values are obtained using paired sample t test, p values < 0.05 is considered to be statistically significant. GI = Gingival Index, PI = Plaque Index, PD = Probing Depth, CAL = Clinical Attachment Level, TAOC = Total Anti-Oxidant Capacity, MDA = Malondialdehyde

# Table 3. Relative % change of values in clinical parameters and biochemical markers in cpgroup (n = 120)

Clinical Parameters	Mean ± SD		
	Relative % Change Values		
GI	38.49 ± 10.31		
PI	38.76 ± 10.33		
PD [mm]	14.52 ± 4.13		
CAL [mm]	20.57 ± 6.07		
Biochemical Markers	Mean ± SD		
	Relative % Change Values		
TAOC [µM/L]	-10.31 ± 2.24		
MDA [nM/ml]	34.71 ± 5.94		

Values are mean ± SD, Relative % change is calculated using the formula [[Baseline value – Post-Treatment value ] / Baseline value x 100], Negative value indicates post treatment higher value and vice versa. . GI = Gingival Index, PI = Plaque Index, PD = Probing Depth, CAL = Clinical Attachment Level, TAOC = Total Anti-Oxidant Capacity, MDA = Malondialdehyde

Clinical Prameters→ Biochemical Markers↓		GI	PI	PD	CAL	
TAOC [µM/L]	r value	.104	220 <sup>*</sup>	452*	045	
	p value	.342	.042	.000	.067	
MDA [[nM/ml]	r value	071	063 <sup>*</sup>	249 <sup>*</sup>	.367**	
	p value	.515	.564	.021	.001	

 Table 4. Correlation of relative % change in clinical parameters and biochemical markers after

 SRP in CP group (n=120)

Values are represented as Pearson Correlation [r values], Negative values indicate inverse association and vice versa, p values are obtained using Pearson Correlation test, p values < 0.05 is considered to be statistically significant. \*. Correlation is significant at the 0.05 level [2tailed]. \*\*. Correlation is significant at the 0.01 level [2-tailed]. GI = Gingival Index, P I= Plaque Index, PD = Probing Depth, CAL = Clinical Attachment Level, TAOC = Total Anti-Oxidant Capacity, MDA = Malondialdehyde

The present study attempts to understand and quantify the relationship between the extent of improvement in clinical periodontal parameters and systemic AO markers such as TAOC and MDA after the SRP therapy is carried out. The associations between CP and OS markers such as TAOC and MDA and the effect of SRP on these markers have been discussed earlier by us [19,23].

The beneficial effects of SRP on systemic inflammatory [21,24] and OS markers [5,22] have also been reported by others. Further the studies [25.26] have been associated with periodontal clinical markers to some biochemical inflammatory [24,27,26] and OS [4,28,29] markers. Some studies [24,25] have shown a positive association between periodontal clinical markers and IL-6, a negative association with IL-10 [30] while others [26,31] couldn't find any association between them. OS markers such as GPx [4], SOD, TAOC [26], and AO potential [32] were also significantly [28] and non-significantly [32] correlated to the periodontal clinical markers.

The study by Teles FR et al 2012 [28] focused on the relationship between IL-6, TNF- $\alpha$ , adipokines, and vitamin D in serum of the CP individuals. They reported a correlation in the serum parameters but found no association among themselves or other clinical parameters [26].

Most of the above studies have been taken into consideration to determine the average or mean values of the study parameters for their correlation and the subsequent conclusions. However, the association between the extent of improvement [relative % change] in the clinical and biochemical parameters has not been reported in the present study. In this study, the mean relative % change values were observed in the CP group for clinical parameters that ranged between 14.52% to 38.76% and those for TAOC & MDA, the values were recorded as -10.31% to 34.71% respectively [Table 3]. Post treatment, TAOC showed higher values as compared to the baseline showing negative values for relative % changes. On the other hand, MDA and other clinical parameters showed lower values post treatment showing positive values for the relative % change.

The correlation between clinical parameters and biochemical markers has been displayed in [Table 4]. The obtained results of this study for TAOC relative % change showed a significant [p ≤ 0.05] negative association with the clinical parameters. For CAL values, the correlation reached to a significant level [p = 0.067]. The MDA showed significant [ $p \le 0.05$ ] correlation to PD and CAL. Furthermore, among the clinical parameters, PD and CAL's relative % change values [14.52% and 20.57% respectively] were relatively lower than those of GI [38.49%] and PI [38.76%] respectively. This indicates that SRP and oral hygiene maintenance may prevent plaque and inflammation but the therapy regime may contribute less in the periodontal tissue building and gaining attachment level, at least through the conditions explored in this study.

Oral clinical improvement is reflected in the systemic improvement. However, as compared to the clinical parameter, TAOC showed lower relative % change values (-10.31% v/s 14.52% to 38.76%, Table 3). While the MDA showed relative % change value competent to those of the clinical values (34.71% v/s 14.52% to 38.76%, Table 3). The findings associated with TAOC may be attributed to the time period of the study (a follow-up after 3 months period) as

biochemical markers may need longer time to show the equivalent improvement as compared to those of the clinical parameters.

biochemical Furthermore, TAOC is а OS marker and a putative index of AO capacity including AO enzymes, metal ion seque strates, vitamins, and other metabolites [33]. It is in a dynamic state and may also be affected by other physico-chemical, environmental, or pathological agents [34]. The monitoring of those variations was beyond the scope of the present study. On the other hand, MDA, a potential biomarker of OS and lipid peroxidation, is highly stable and hence may have shown a differential response as compared to TAOC. Albeit varying response, it is promising to note that SRP was effective in improving clinical and biochemical markers as compared to their corresponding baseline values.

Also, the extent of improvement (the relative % change) in clinical parameters showed some correlation to the improvement in biochemical markers, although the correlation could not reach statistical significance for all the parameters such as GI, CAL for TAOC, and GI for MDA.

## 4. CONCLUSION

Hence, the scaling and root planning (SRP) offers promising improvements in clinical and biochemical parameters. The SRP therapy can reduce local inflammation and improve the systemic health in individuals with periodontitis. Because of these improvements, the dental professionals may include the SRP therapy as a beneficial tool in their therapy regime. The relative % change may be used as a possible quantifiable measure for the assessment of the therapy outcome.

## CONSENT

"All authors declare that 'written informed consent was obtained from the patients for publication of this manuscript."

## ETHICAL APPROVAL

"All authors hereby declare that all the experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki."

### **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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