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Evaluation of efficacy of locally delivered Herbal extract in treatment of Chronic periodontitis –A Randomised controlled trial

Research Article

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Abstract

Background: Various locally delivered agent are used as an adjunct to scaling and root planning (SRP). *8-Hydroxydeoxyguanosine (8-OHdG)* is best documented biomarker of oxidative stress in periodontitis. Objective: To evaluate the efficacy of locally delivered *Ocimum sanctum gel* by assessing the levels of 8-OHdG in Gingival crevicular fluid (GCF) of smokers and nonsmokers with periodontitis. Material and Methods: The study included 50 patients divided into 3 groups, group I - periodontally healthy, group II- smokers with stage II periodontitis and group III- non-smokers with stage II periodontitis. Smokers and non-smokers with stage II periodontitis received the locally delivered 10% OS gel as an adjunct to SRP at the test site at baseline and after 1 week recall, while SRP alone at control site. Result: 8-OHdG GCF levels were analyzed with enzyme-linked immunosorbent assay (ELISA) at baseline, and 6-months follow up. Probing pocket depth (PPD) in test site significantly reduced to 2.75 ±0.64, 2.50 ±0.68 at 6 month after periodontal therapy in group 2 and 3 respectively. Similarly, clinical attachment level (CAL) in test site significantly reduced to 3.15 ±0.81, 2.85 ±0.93 at 6 month after periodontal therapy in group 2 and 3 respectively. Conclusion: The application of 10% *Ocimum sanctum* L. *gel* showed significant improvement in PPD, CAL and PI and GI and reduction in GCF 8-OHdG levels.

Key Words: 8-hydroxyguanosine, Gingival crevicular fluid, Local drug delivery, *Ocimum sanctum*, Oxidative stress, Reactive oxygen species.

Introduction

Periodontitis occurs refers as a result of complex interaction between the pathogenic bacteria and host immune response, (1) accumulation of plaque and calculus, the proliferation of pathogenic organisms residing subgingival sulcus and long-term release of reactive oxygen species (ROS) and neutrophil enzymes which leads to progressive attachment loss, bone loss, inflammation and destruction of the elements of the periodontium. (2)

Excessive production of ROS cause various cellular and extracellular tissue damage by DNA oxidation, lipid peroxidation, protein disruption and proinflammatory cytokine induction.(3) In periodontitis the infiltration of neutrophil, fibroblast, osteoclasts & endothelial cells predominantly lead to an increase ROS level resulting break down of epithelium and damage to connective tissue in the proximate area.(1) In the oral cavity, free radicals can result from external sources, such as nicotine.(4) Nicotine is main component in tobacco smoke and has the potential to inhibit neutrophil, ROS release at low doses, impairing the elimination of periodontal pathogens and at high dose nicotine stimulates ROS release that causes oxidative stress mediated tissue damage in periodontal tissue. Thus, tobacco smoke exaggerates the severity of periodontal destruction. (5) However, oxidative stress is reversed as free radicals are neutralized through the presence of antioxidants.

8- Hydroxydeoxyguanosine (8-OHdG) is biomarker of oxidative stress in various diseases including periodontitis. So, increased 8-OHdG level can be considered a sign of oxidative DNA damage of periodontal tissues. (6)

Local drug delivery with antimicrobial was initiated to improve nonsurgical therapy by aiding as an adjunct to scaling and root planing. Various studies reported the efficacy of the local drug delivery (LDD) systems and found to be effective in the treatment of periodontitis. (7,8) But most of these LDD systems are expensive and difficult to procure. Undesirable side effect such as gastrointestinal disturbances and development of antibiotic resistance and occur by the

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use of these LDD. Several research studies have shown the beneficial use of herbal drugs like aloe vera, neem, propolis, cranberry, turmeric, ocimum sanctum etc. in the treatment of periodontal diseases. (9, 10, 11)

Ocimum sanctum is reported to have antibacterial and anti-inflammatory properties and effective in neutralizing halitosis, also vitamin A and C, calcium, zinc, iron, chlorophyll and many other phytonutrients contained in ocimum sanctum are scavenging antioxidants. (5,12,13)

As here is paucity of literature on combined used of ocimum sanctum with Scaling and root planing in the treatment of stage II periodontitis, the present clinical trial aims to evaluate and compare the efficacy of locally delivered *ocimum sanctum gel* and *8-OHdG level* in GCF in smokers and nonsmokers with stage II periodontitis.

Material and methods

Study design

This split mouth randomized controlled clinical trial included 50 patients with the age range of 30-55 years visiting the department of periodontology of our Institute. After being approved by the Institutional Ethics Committee the study was done in accordance with the Helsinki Declaration of 1975, as revised in 2013 (approved no ECR/885/Inst/MH/2017). The present study was registered at clinical Trial Registry–India, (CTRI/2017/10/015721) being primary register of the WHO International Clinical Trials Registry Platform. Prior to the initiation of the study a written informed consent was obtained from all the patients willing to participate in the study. Smokers were enrolled if they smoked 10 cigarettes/day, and non-smokers were considered as not having smoked cigarettes in their lifetime.

Participants

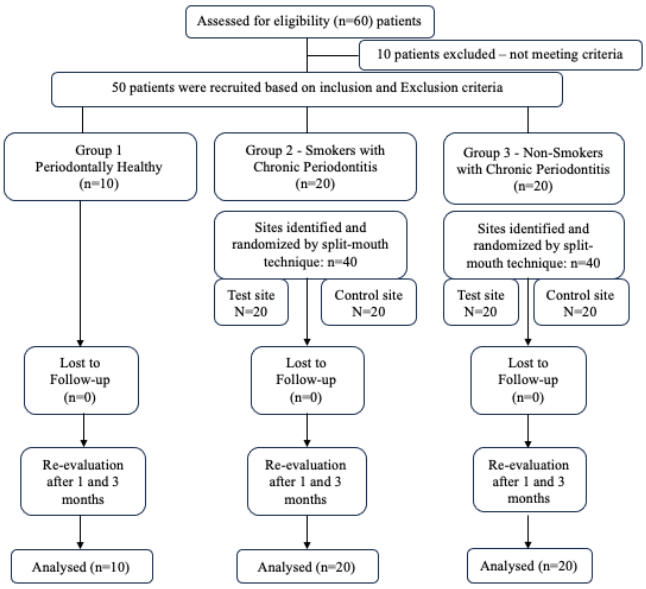
The patients were divided into three groups. Group 1 comprised of 10 periodontally healthy patients, Group 2 consisted of 20 smokers with stage II while Group 3 included 20 non-smokers with stage III.

The recruitment process is illustrated in the study flow chart (Fig No. 1)

Intervention

Scaling and root planing was performed by single operator and oral hygiene instructions were given to the patients. The proposed study uses Ocimum sanctum gel as an adjunct to SRP in patients with stage II periodontitis. This split mouth study with two interventions to be randomly assigned to each site in the same patient. The Tulsi gel was injected into the site with periodontitis into the pocket with the help of a 2ml syringe the gel was filled until the pocket is full of the gel and spills off the margins of the pocket. Patient was recalled after 1 week for re-administration of 10 % ocimum sanctum gel. Control site received SRP alone in Group 2 and Group 3 at baseline. The patients were re-evaluated for PI, GI, PPD and CAL after 1 and 6 months by same calibrated examiner.

Fig No.1: Flow Chart of the Study



Outcomes

The primary outcomes measures were the changes in the levels of 8-OHdG level in GCF in smokers and non-smokers with stage II periodontitis. The change in the clinical parameters like Plaque Index (PI),(14) Gingival Index (GI) (15) Probing pocket depth (PPD) and Clinical attachment level (CAL) were measured at baseline, 1 and 6 months for the secondary outcome measures.

Sample size

With reference to the article by Chandra et al., (1) in which the author evaluates the efficacy of local drug delivery system of lycopene gel as an adjunct to scaling and root planing for the treatment of human periodontal defects. A 1 % difference between the two groups in the mean PPD and standard deviation of 2.5 was considered from the study. The confidence interval was set at 95% (P= 0.05). Sample size was achieved by using G power 3.1 statistical power analysis software. A total number of 50 patients including 20 smokers and 20 non-smokers would provide the 80%Power (β= 0.2) (1) and hence recruited in the study.

Randomization

In this split mouth study each patient's upper or lower arch were randomly divided into test and control site on the basis of coin toss method. Total 80 sites with PPD (≥ 5 mm) were assigned as Test site and received scaling and root planing along with application of 10 % Tulsi gel.

The study included test and control group. After assigning the patients, the same investigator (SR) provided the prescriptions. All patients were treated by another investigator who was blinded to the designation of the groups and was not aware of which subjects were receiving the gel. This investigator provided the clinical treatment modalities, measurements, and sampling. The treatment codes of the study were not available to the treating investigator (SM) until the data were completely analyzed by the statistician

For the inclusion criteria Periodontally healthy patients, Untreated periodontitis patient where diseased site had probing pocket depth ≥ 5 mm and clinical attachment level (CAL) ≥5mm, Patient who were current smokers and with history of smoking approximately 10 cigarettes per day from last 3 years.

Patients with any systemic disease, history of acute illness, pregnant, postmenopausal or lactating women, who had undergone any type of periodontal therapy or oral prophylaxis in past 6 months, or those who received antibiotic, anti-inflammatory drugs, antioxidant, vitamin, alcohol during study period were excluded from the study.

Periodontal examination

In first visit, a written informed consent was signed by the patients, then a complete history of the selected patients was recorded followed by clinical and radiographic examination. At least two contralateral periodontal pockets were selected from each patient. Clinical parameters including Plaque Index (PI), (14) Gingival Index (GI) (15) were measured at baseline, 1 and 6 months. Probing pocket depth (PPD) and Clinical attachment level (CAL) was recorded at baseline, 1 and 6 months with the help of periodontal probe (UNC 15) and the measurements were standardized using custom-made acrylic stent. All clinical parameters were assessed by single calibrated examiner (SM) at least thrice and the mean value was taken into consideration.

Intra-class correlation (ICC) coefficient with two-way mixed effects model were obtained for GI, PI, and PPD, CAL. The ICC ranged 0.86 to 0.92 in the groups ($p < 0.0005$), indicating good intra-observer reliability.

Material For Gel Preparation

- Carbopol
- Water
- magnetic bead
- 10 % of tulsi extract
- sodium benzoate
- methyl paraben
- propyl paraben

Preparation of 10% ocimum sanctum (OS) Gel (16)

For preparation of 10% ocimum sanctum gel, Carbopol 1000 mg soaked with 50 ml of water in a beaker was kept for a day. Next day the beaker was placed on magnetic stirrer, set at 1000 RPM followed by addition of magnetic bead and stirred for two hours. Later, 10 % of tulsi extract (Supercritical fluid extract, procured from Sami labs, Bengaluru) was added and stirred followed by addition of sodium benzoate 25 mg, methyl paraben 10 mg, propyl paraben10 mg and a little quantity of water. The rotations per minute (RPM) was changed to 1500 and the contents are stirred for eight hours with addition of water intermittently as and when required. After obtaining perfect gel consistency, it was transferred and dispersed in a plastic container. Prepared gel was exposed under Ultra Violet light in UV chamber for a period of 30 minutes. The gel integrity was checked. Methyl paraben and propyl paraben were added as preservatives which prevent the growth of bacteria and keeps the gel stable.

Baseline measurement and application of gel

GCF sample collection

GCF samples were collected using microcapillary pipette and in the morning following an overnight fast of 8 hours, during which patients were instructed not to drink (except water) or eat. GCF sample was collected from the selected sites with maximum attachment loss ((CAL) ≥ 5 mm) for CP in Group 2 and Group 3 and from a non-diseased site in Group 1 individuals. After isolating the site with cotton rolls, plaque along with the supragingival calculus was removed using area-specific Gracey curette ‖ and cotton gauze, to avoid contamination and blocking of microcapillary pipette § by plaque. The sulcular areas were gently air-dried. A color-coded 1–5 µl, calibrated volumetric microcapillary pipette § was placed at the entrance of the gingival crevice until a standardized volume of 2 µl of GCF was collected. The GCF samples which were contaminated with blood or saliva and air bubbles were discarded, and fresh samples were collected (17). Collected GCF samples were immediately transferred to airtight plastic vials (Eppendorf tubes) and stored at -30 °C until final assayed.

Determination of GCF 8-OHdG by ELISA

GCF, 8-OHdG levels were assessed using enzyme-linked immunosorbent assay (ELISA) (Krishgen Biosystems Human ELISA kit, Mumbai, India) according to the manufacturer's instructions.

Statistical analysis

Statistical software (SPSS ver 24.0 [IBM] Corp]) was used for data analysis. Intragroup comparison of PPD, CAL, GI and PI between various groups was performed using repeated measures analysis of variance (ANOVA) followed by multiple comparison test Bonferroni correction. GCF 8-OHdG levels were measured among different groups using paired t- test, p value <0.05 was considered to be statistically significant. Intergroup comparison of clinical parameters and between various groups were performed using paired t- test.

Result

The descriptive statistics for PI and GI of patients in all the three study groups expressed as the mean and standard deviation of periodontal parameters are depicted in Table No. 1. PI in test site was 2.14 ±0.37, 2.05 ±0.29 at baseline which reduced significantly to 0.72 ±0.28, 0.56 ±0.21 after periodontal therapy in group 2 and group 3 respectively. Similarly, GI in test site was 1.91 ±0.38, 2.28 ±0.44 at baseline which reduced significantly to 0.68 ±0.28, 0.45 ±0.17 after periodontal therapy in group 2 and group 3 respectively. Likewise, control sites also showed significant improvement in PI and GI values after periodontal therapy in all the three groups.

The descriptive statistics for PPD and CAL of patients in all the three study groups expressed as the mean and standard deviation of periodontal parameters are depicted in Table No. 2. PPD in test site was 5.50 ±1.00, 5.30 ±0.57 at baseline which highly significantly reduced to 2.75 ±0.64, 2.50 ±0.68 at 6 months after periodontal therapy in group 2 and group 3 respectively. Similarly, CAL in test site was 5.50 ±1.00, 5.30 ±0.57 at baseline which highly significantly reduced to 3.15 ±0.81, 2.85 ±0.93 at 6 months after periodontal therapy in group 2 and group 3 respectively. Also control sites showed significant improvement in PPD at 6 months, but CAL showed non-significant changes at 6 months after periodontal therapy in group 2 and group 3 respectively.

Table No. 3 provides descriptive statistics for 8-OHdG level in GCF in patients from the three study groups. 8-OHdG values in Test sites were 10.40 ±1.94 ng/ml, 8.20 ±1.63 ng/ml at baseline which highly significantly reduced to 6.06 ±1.67 ng/ml, 4.42 ±0.57 ng/ml at 6 month post therapy in groups 2 & 3 respectively. Similarly control sites showed 10.25 ±1.59 ng/ml and 8.14 ±2.13 ng/ml at baseline which significantly reduced to 8.29 ±1.38 ng/ml and 7.13 ±1.97 ng/ml at 6 months post therapy in group 2 and 3 respectively.

Table No. 4 provides intergroup comparison of PI, GI, PPD, CAL 8-OHdG levels in test and control sites in group 2 and group 3. A positive correlation was found in PPD, CAL and 8-OHdG level in test as compared to control site with $p< 0.005$. Table No. 5 shows that 8-OHdG levels were significantly higher in group 2 compared to group 3 in test as well as control site at baseline and 6 months.

Table No.1: Intra group Comparison of PI and GI among the study groups at different time interval

	Plaque Index Mean± SD				**Gingival index Mean±SD**			
Groups	Base line	1 month	6 month	P-value	Base line	1 month	6 month	P-value
Group 1 **(Healthy)**	0.61 ±0.24	0.41 ±0.16	0.58 ±0.50	P = 0.003*	0.36 ±0.24	0.27 ±0.13	0.03 ±0.04	P= 0.001*
Group 2								
Test Site	2.14 ±0.37	1.55 ±0.36	0.72 ±0.28	P=0.000*	1.91 ±0.38	1.36 ±0.35	0.68 ±0.28	P=0.000*
Control Site	2.17 ±0.40	1.62 ±0.36	0.82 ±0.23	P=0.000*	1.91 ±0.30	1.46 ±0.25	0.88 ±0.37	P=0.000*
Group 3								
Test Site	2.05 ±0.29	1.32 ±0.24	0.56 ±0.21	P=0.000	2.28 ±0.44	1.11 ±0.45	0.45 ±0.17	P=0.000
Control Site	2.25 ±0.40	1.32 ±0.24	0.98 ±0.28	P=0.001*	2.27 ±0.44	1.32 ±0.32	0.58 ±0.24	P=0.000*

*Mean value Significant measured by Repeated measures ANOVA
Data is expressed as mean±SD. GI– Gingival index; PI – Plaque index; SD – Standard deviation

Table No. 2: Intra group Comparison of PPD and CAL among the study groups at different time interval

	PPD Mean± SD				**CAL Mean±SD**			
Group	Base line	1 month	6 month	P-value	Base line	1 month	6 month	P-value
Group I **(Healthy)**	2.10 ±0.99	1.20 ±0.91	0.40 ±0.51	P= 0.027*	2.10 ±0.99	1.90 ±0.73	0.20 ±0.78	P=0.008
Group II								
Test Site	5.50 ±1.00	4.10 ±0.72	2.75 ±0.64	P=0.000*	5.50 ±1.00	4.20 ±0.77	3.15 ±0.81	P=0.000*
Control Site	5.30 ±0.73	5.30 ±0.73	5.10 ±0.78	P=0.042*	5.30 ±0.73	5.25 ±0.71	5.10 ±0.85	P=0.232
Group III								
Test Site	5.30 ±0.57	4.00 ±0.56	2.50 ±0.68	P=0.000*	5.30 ±0.57	4.15 ±0.50	2.85 ±0.93	P=0.000*
Control Site	5.25±0.91	4.75 ±0.97	4.20 ±0.95	P=0.001*	5.30 ±0.92	5.00 ±0.85	4.70 ±1.08	P=0.004*

*Comparison of Mean value Significant measured by Repeated measures ANOVA
Data is expressed as mean±SD; PPD- Probing pocket depth; CAL – Clinical attachment level

Table No. 3: Intra group Comparison of GCF 8-OHdG Levels among the study groups at baseline and 6 months

	GCF 8-OHdG Levels**(ng/ml)**		
Group	Base line	6 month	P-value
Group I	4.44 ±0.48	4.27 ±0.33	P= 0.222
Group II			
Test Site	10.40 ±1.94	6.06 ±1.67	P=0.000*
Control Site	10.25 ±1.59	8.29 ±1.38	P=0.001*
Group III			
Test Site	8.20 ±1.63	4.42 ±0.57	P=0.000*
Control Site	8.14 ±2.13	7.13 ±1.97	P=0.002*

*Comparison of Mean value Significant measured by paired t test.
Data is expressed as mean ± SD; GCF – gingival crevicular fluid;8-OHdG- 8 hydroxyguanosine

Table No. 4: Inter group comparisons of PI, GI, PPD, CAL and 8-OHdG levels between Test and Control sites of Group 2 and Group 3

	Group 2 (Test site vs control site)		**Group 3 (Test site vs control site)**	
Clinical Parameter	**t**	**Sig.**	**t**	**Sig .**
PI Baseline	-0.207	0.837	-1.795	**0.081**
PI 1 Month	-0.659	0.514	0.000	**1.000**
PI 6 Month	-1.350	0.185	-5.414	**0.000***
GI Baseline	0.000	1.000	0.000	**1.000**
GI1Month	0.000	1.000	-1.695	**0.098**
GI 6 Month	0.000	1.000	-2.080	**0.044***
PPD Baseline	0.827	0.414	0.000	**1.000**
PPD 1 Month	-2.414	.021*	-6.296	**0.000***
PPD 6 Month	-5.659	.000*	-11.113	**0.000***
CAL Baseline	0.657	0.515	0.000	**1.000**
CAL 1 Month	-3.107	.004*	-5.670	**0.000***
CAL 6 Month	-5.126	.000*	-7.962	**0.000***
8-OHdG Baseline	0.266	0.791	0.100	**0.921**
8-OHdG 6 Month	**-4.592**	**.000***	**-5.880**	**0.000***

*Statistical significance measured by paired T test. Data is expressed as mean±SD. GI– Gingival index; PI – Plaque index; PPD- Probing pocket depth; CAL – Clinical attachment level; 8-OHdG- 8 hydroxyguanosine; SD – Standard deviation

Table No. 5: Inter group comparisons of PI, GI, PPD, CAL and 8-OHdG levels between Group 2 And Group 3 with chronic periodontitis in Test

	Test site **(Group 2 Vs Group 3)**		Control site **(Group 2 Vs Group 3)**	
Clinical Parameter	t	Sig.	t	**Sig.**
PI Baseline	0.908	0.369	-0.834	**0.410**
PI 1 Month	2.416	0.021*	2.548	**0.015***
PI 6 Month	2.032	0.049*	-1.744	**0.090**
GI Baseline	-2.832	0.007*	-2.006	**0.052***
GI 1Month	1.920	0.062	0.934	**0.357**
GI 6 Month	3.222	0.003*	1.494	**0.144**
PPD Baseline	0.777	0.442	-0.728	**0.471**
PPD 1 Month	-2.280	0.028*	0.490	**0.627**
PPD 6 Month	-3.231	0.003*	1.191	**0.241**
CAL Baseline	0.777	0.442	-0.522	**0.605**
CAL1 Month	0.246	0.807	-1.206	**0.235**
CAL 6 Month	1.084	0.285	-1.520	**0.137**
8-OHdG Baseline	3.869	0.000*	3.116	**0.004***
8-OHdG 6 Month	**4.135**	**0.000***	**2.063**	**0.046***

*Statistical significance measured by paired TtestData is expressed as mean ± SD. GI– Gingival index; PI – Plaque index; PPD- Probing pocket depth; CAL – Clinical attachment level; 8-OHdG- 8 hydroxyguanosine; SD – Standard deviation

Discussion

Oxidative stress is characterized by an imbalance between free oxygen radicals and antioxidant defense system and is capable of causing damage to various cellular and extracellular components. [1] The oxidative damage generally occurs after exposure to a high concentration of ROS and/or a decrease in antioxidant defense system against ROS. Oxidative stress has been implicated as a major contributor in over 100 disorders and more recently periodontitis. (16) Existent literature is in dearth of knowledge regarding evaluating the efficacy of 10% ocimum sanctum gel in periodontal therapeutics and hence a direct comparison with previous studies is quiet perplexing.

In the present study, we assessed oxidative stress markers in GCF of smokers and non-smokers with stage II periodontitis undergoing initial periodontal therapy. It was found that after SRP and LDD, 8-OHdG levels in GCF significantly decreased in both periodontitis groups. All the three groups showed significant reduction in mean Plaque index (PI), Gingival index (GI), PPD and CAL from baseline to 6 months in both sites i.e. test and control sites after phase I therapy statistically significant differences were observed in all groups at different intervals. Also, mean GI showed significant reduction after periodontal therapy in all three groups when compared at baseline to 6 months. This result is in accordance with study done by Chandra et al. (1) where a significant reduction in PI, GI, PPD and CAL was found after periodontal therapy amongst smokers and non-smokers. Similar results were found in some studies showing significant reduction in PI, GI, PPD and CAL after initial periodontal therapy. [6,17,18] The mean PPD was significantly reduced after 1 and 6 months follow up as compared to the baseline in both test & control sites in group 2 and group 3 however, it is important to note that in Test sites, pocket depth reduction was significantly higher than compared to control site. It could be attributed to the anti-inflammatory activity of 10 % ocimum sanctum gel and the variable amount of linoleic acid present which has capacity to block both cyclooxygenase and lipoxygenase pathway of arachidonate metabolism also inhibiting the bacterial growth thus decreasing the formation of plaque. (19,20)

J Gaur et al. revealed that subgingival irrigation with 4% Ocimum sanctum may be effective in reducing plaque accumulation, gingival inflammation and bleeding and has no adverse effects as compared to chlorhexidine. (20) M Hosamane et al., reported that ocimum sanctum extract showed inhibition of periodontal pathogen in vitro and in vivo studies and that there was no statistical difference between holy basil and chlorhexidine mouth rinse with respect to PI. (22) In vitro studies done by S Mallikarjun et al. showed 5 % and 10 % concentration of tulsi extracts demonstrated antimicrobial activity against A. actinomycetemcomitans similar to doxycycline. (23)

In the current study, we found positive correlation between 8-OHdG level in GCF of group 2 and group 3. Following SRP, 8- OHdG level in GCF significantly decreased in both periodontitis groups. This finding is in accordance with previous literature. (18,24) Mechanical removal of plaque helps resolve inflammation and remove periodontal pathogens which results in reduction of oxidative stress. (6) Sezer et al. found that mean 8-OHdG level in the saliva of the CP group were significantly higher than healthy and chronic gingivitis groups. Statistically significant correlation was observed between the salivary levels of 8-OHdG and probing depth (PD) and CAL with ($p < 0.001$) in CP group. (25) Hence the authors suggested that elevated salivary levels of 8-OHdG may be a marker for current disease activity, and it may reflect indirectly disease severity parameters. In present study, intergroup comparison was made for 8-OHdG level, and it was evident that there was a significant reduction in Group 2 as compared to Group 3 with a p value <0.05. This may be due to improved oral hygiene and antioxidant and anti-inflammatory effect of ocimum sanctum gel directly on diseased site. Teruo Inoue et al. reported the 8-OHdG level was significantly higher in smokers than non-smokers, which were significantly reduced by the smoking abstention for 4 weeks as well as smoking abstention with vitamin C supplementation (2g/day) group. (26) Fraga et al. stated that a high intake of vitamin C protected against 8-OHdG formation in human seminal DNA. (27)

A Recent systematic review on herbal extract stated that though the different chemical adjunctive products showed standard results treatment of periodontitis, Herbal extracts like A. Vera shows promising results in reducing plaque and GI scores, without reported side effects. (28)

However, the study has certain limitations as the selection of the patients was made on the basis of clinical indicators which might not necessarily reflect active periodontal destruction. Another limitation of the study is small sample size. Due to the low prevalence of female smoker in India, the group 2 include mostly male patients. It has been suggested that the estimation of serum or saliva or GCF cotinine assays is more reliable for the evaluation of smoking status.

The present study suggests that inflammatory reaction reflects involvement of oxidative stress in biologic fluid in periodontal environment therefore it has been concluded that elevated OHdG levels should be taken as evidence of impaired DNA repair as a result of reactive oxygen species. The mechanism behind these result offers an attractive topic for future studies.

Conclusion

Within the limitations of the present study and to the best of author's knowledge following conclusion can be drawn that the use of Ocimum sanctum 10% gel as an adjunct to SRP is more effective than SRP alone to treat periodontal disease. GCF 8- OHdG level may be a one of the diagnostic and prognostic marker for assessing the severity of periodontal disease and efficacy of periodontal therapy.

Footnotes

‡ PCPUNC 15; Hu Friedy, Chicago, IL,USA.

§ Labo Glass Scientific Supply Co. Haryana, India

‖ Area-specific Gracey curette, Hu Friedy, Chicago, IL,USA.

¶ Krishgen Biosystems Human 8-Hyroxydeoxygunosin (8-OHdG) ELISA K12-1560, Mumbai, India.

SPSS ver 24.0 (IBM Corp.)

Conflicts of interest

The authors report no conflicts of interest related to this study. The study was self-funded by the authors and their institution.

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