Original Article

Pharmacodynamic Potential of Sub-Gingivally Delivered Lovastatin in The Treatment of Patients with Chronic Localized Moderate Periodontitis: A Longitudinal Study

Hejal K. Bhamtikar [1], Bindu R. Nayar [2], Lalbahadur L. Sadhu [3], Aayushi Gupta [4, 5], Mainul Haque [6, 7]

ABSTRACT

INTRODUCTION

Periodontitis is an inflammatory disease caused by specific bacteria that leads to the progressive destruction of tooth-supporting tissue. Treatment often includes non-surgical therapy, but antimicrobials may be needed to target bacteria. However, antimicrobial resistance can occur. Statins, particularly lovastatin, have shown promise in reducing inflammation and promoting bone formation in periodontitis treatment.

Methods

This study included 62 patients (12 males, 50 females) with chronic localized periodontitis aged 29-41. Patients were randomly divided into two groups: one treated with lovastatin gel (experiment group) and the other with a placebo (control group). After baseline measurements, subgingival scaling and gel application were performed. Data was analyzed using SPSS software (IBM SPSS Statistics for Windows, Version 22.0.Armonk, NY: IBM Corp.).

Results

This longitudinal study evaluated the efficacy of subgingival lovastatin in treating chronic localized moderate periodontitis. The study included 62 patients with no significant baseline differences. Significant improvements were observed in the lovastatin-treated experimental group for sulcus bleeding, pocket probing depth, clinical attachment, and bone fill (93.5% vs. 29% in the control group)

Conclusion

The study concludes that lovastatin's local delivery adjunct to scaling and root planning reduces bleeding pocket depth and enhances clinical attachment and bone growth. Long-term, more extensive studies are needed to assess its full benefits.

Keywords

beneath the gums, subgingival, local drug delivery, mouth and tooth diseases, chronic periodontitis, effectiveness, therapeutic intervention, a fungal metabolite, lovastatin

INTRODUCTION

Periodontitis is an inflammatory disease of the tooth-supporting tissue of the teeth caused by specific microorganisms resulting in progressive destruction of the periodontal ligament with pocket formation, recession, or both [1,2]. Inflammatory periodontal disease is widely accepted and caused by dental plaque bacteria [1]. The severity of periodontitis mainly depends on the host's response to the bacterial invasion [3]. The need to quantify the specific bacteria in periodontal disease started in the 1970s and has become a prominent area of investigation. The concept of the plaque hypothesis and

1. Department of Periodontology, NAMO Medical Education and Research Institute, Shri Vinoba Bhave Civil Hospital, Daman and Diu, Dadra and Nagar Haveli, India.
2. Department of Periodontology, Government Dental College Thiruvananthapuram, Kerala, Thiruvananthapuram, India.
3. Department of Oral Pathology and Microbiology, Ahmedabad Dental College and Hospital, Gujarat, Ahmedabad, India.
4. Department of Periodontology, Karnavati School of Dentistry, Karnavati University, Gujarat, Gandhi Nagar, India.
5. Consultant Data Analyst, Business Ops, Natera Inc, Texas, USA.
6. Department of Pharmacology and Therapeutics, Faculty of Medicine and Defence Health, National Defence University of Malaysia, Kuala Lumpur, Malaysia.
7. Department of Research, Karnavati School of Dentistry, Karnavati University, Gujarat, Gandhinagar, India

Correspondence

Mainul Haque. Unit of Pharmacology, Faculty of Medicine and Defence Health, Universiti Pertahanan Nasional Malaysia (National Defence University of Malaysia), Kem Perdana Sungai Besi, 57000 Kuala Lumpur, Malaysia. **Email**: runurono@gmail.com, mainul@upnm.edu.my. Cell Phone: +60109265543

DOI: https://doi.org/10.3329/bjms.v24i2.81529

different forms of periodontal diseases are caused by specific bacteria [2,4,5]. Non-surgical therapy is often the preferred choice of treatment for periodontal disease. Still, it may not eliminate the anaerobic infection at the bottom of the periodontal pocket as it is inaccessible to the periodontal instruments [6]. The remaining bacteria at the base of the pocket often recolonize, causing an inflammatory state [7].

So, researchers came up with the idea of adding antimicrobials to local and systemic treatment protocols. This served as an adjunct to mechanical therapy. Systemic antimicrobials helped eliminate the bacteria left over after mechanical treatment [8]. However, the above advantage comes with certain disadvantages, such as the risk of antibiotic resistance and adverse effects, which limits its age [9]. In 1979, Max Goodson developed a local drug delivery therapeutic agent. This system overcame most systemic therapy problems, limiting the drug to its target site and achieving a much higher concentration [10].

Various antimicrobials have been tried for local delivery in the subgingival areas, e.g., tetracycline, chlorhexidine, metronidazole, minocycline, azithromycin, doxycycline, and ofloxacin [11]. Local drug delivery can be used in gels, microspheres, etc. Multiple vehicles like methylcellulose, Carbopol, and glycolide polymers have been used and mentioned in the literature [12-14]. Similarly, various studies have been conducted on the different modes of local drug delivery of the antimicrobial subgingivally [15]. The local use of antibiotics can lead to chances of resistance to microorganisms, hence reinfection from the neighboring sites like the tongue, buccal mucosa, etc. [16]. Thus, developing an agent that can heal that issue without antibiotic resistance can be of great utility for managing chronic localized moderate periodontitis.

Various studies have shown that statins effectively treat periodontitis by reducing inflammation, promoting bone formation, and reducing tooth loss in chronic periodontitis. In addition, the literature has shown the possible effects of statins on the local inflammatory response associated with periodontal disease. Individuals taking statin drugs experience significantly less periodontal inflammation, as measured by bleeding on probing, despite equivalent plaque levels, PPD, and smoking status.

Hence, this study assessed the efficacy of subgingivally delivered lovastatin in treating patients with chronic localized moderate periodontitis.

MATERIALS AND METHODS

This longitudinal study (LS) was conducted in the Department of Periodontology, Ahmedabad Dental College & Hospital, Ahmedabad, Gujarat 382115, India. It aimed to compare the efficacy of subgingivally delivered lovastatin as an adjunct to scaling and root planning in treating patients with chronic localized moderate periodontitis. "LSs employ continuous or repeated measures to follow individuals over prolonged periods—often years or decades. They are generally observational, with quantitative and/or qualitative data collected on any combination of exposures and outcomes, without any external influence being applied" [17]. No attempt was made to do a randomized control trial (RCT).

Sample Size

The study included 62 patients (12 males and 50 females) aged 29 to 41, with a mean age of 33.6 years in the test group and 35.1 years in the control group. Patients of either sex, systemically healthy, with chronic localized periodontitis, one or two sites involved with attachment loss, pocket probing depth of 4 to 7 mm confirmed by radiograph, and no tooth mobility were included in the study. The following formula was used to determine the sample size.

[n=\frac{(Z_{\alpha/2} + Z_{\beta})^2 \cdot 2 \cdot \sigma^2}{d^2} \] (Where Z α/2 is the critical value of the Normal distribution at α/2).

Patients allergic to the statin group, smokers and Alcoholics, those with uncontrolled systemic conditions, pregnant and lactating females, and those undergoing periodontal treatment in the last 6 months were excluded from the study.

Study Period

This study was carried out from August 2021 to February 2023.

Study Design

This is a single-blinded, prospective study. All 62 patients were randomly divided into 2 groups of 31 each (Figure 1). The experimental group consisted of 7 males and 24 females, and the control group consisted of 5 males and 26 females. The randomization procedure was done using the coin toss method. Group One consisted of the patients treated with lovastatin gel, whereas group 2 patients were treated with carbopol gel 980 [control (placebo) group].

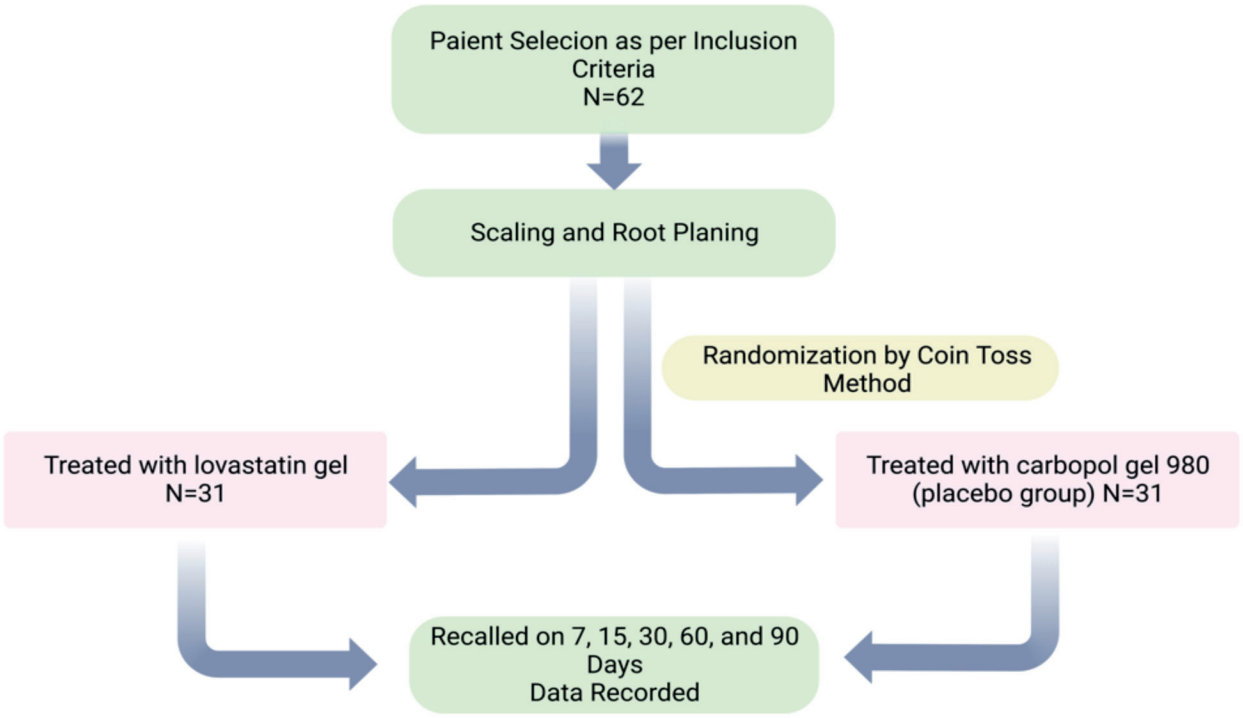

Figure 1: Flowchart illustrating the methods of this paper.

Notes: This illustration was created using the premium version of Biorender [18] (https://www.biorender.com/), with the License Agreement number NA27MVV5JM, accessed on December 6th, 2024.

Illustration Credit: Aayushi Gupta.

Treatment Procedure

Patients with chronic localized moderate periodontitis were selected. All the patients underwent supra-gingival scaling and were recalled after one week. Baseline measurements like plaque Index [(PI), Löe, 1967] [19], gingival index [(GI), Turesky-Gilmore-Glickman] [19-21], probing pocket depth (PPD) [21,22], modified sulcus bleeding index [(mSBI) by Mombelli, 1987] [21,23], clinical attachment level [24], and an intraoral periapical (IOPA) radiograph [25,26] were taken for all the patients. The IOPA was taken using a calibrated grid and paralleling cone beam technique. Soon after recording the baseline measurements, patients were randomly assigned to the test or the control group using coin toss methods. All the patients underwent subgingival scaling and root planning.

Additionally, group one patients were treated with lovastatin gel (experimental group) and group two with placebo carbopol gel (control group). The gel was placed with the help of a syringe and needle in both groups. Periodontal dressings were placed to protect the sites. Post placement of the gel, patients were given oral hygiene instructions using flip charts and were instructed to refrain from chewing hard or sticky foods, brushing near the treated areas, and using any interdental aids for one week. They were also advised to report any untoward effects. The recall visits were scheduled on the 7th, 15th, 30th, 60th, and 90th day. All the parameters were recorded on each visit. A trained, calibrated single examiner did all the measurements and recordings.

Preparation of Lovastatin Gel (Experimental Group)

A 3 gm Carbopol 980 (Corel Pharma Chem, GJ, India) was weighed using a balance (Shimadzu Ax100 electronic balance, Shimadzu Forest, Japan) and transferred in small quantities to a beaker containing 100 ml of distilled water, which was subjected to continuous stirring using a laboratory scale stirrer (Eltek, Mumbai, India) at 1000 rpm. This mixture added 1.2mg of lovastatin powder (Sterling Biotech

Ltd, GJ, India). Stirring was done Continuously till the uniform gel was obtained. This gel was transferred into previously sterilized, amber-colored glass bottles. The sterilization was done by autoclaving at 125 degrees centigrade for 21 minutes at 15 lbs. No preservatives or flavoring agents were added. The gel was prepared fresh daily and stored at an ambient temperature for use within 24 hours.

Preparation of Carbopol Gel (Control Group)

Carbopol powder is typically obtained at 2% w/w and was sprinkled slowly into distilled water while stirring continuously to avoid clumping. The solution was allowed for 2 hours to hydrate appropriately until a uniform solution was obtained. This is a significant step in gel formation. Triethanolamine was added slowly till the pH reached 6.0 to 7.0. After this neutralization, incorporate preservatives, active ingredients, or other additives, ensuring they are thoroughly mixed without destabilizing the gel structure. Once the gel is ready, transfer it into clean, sterilized containers to avoid contamination and store it at room temperature.

Statistical Analysis

The data gathered were organized and subjected to statistical analysis using SPSS software (IBM SPSS Statistics for Windows, Version 22.0. Armonk, NY: IBM Corp.). Using the standard probability plot, it was noted that the clinical parameters follow the distribution normality. Therefore, an independent t-test was used for plaque index, bleeding on probing, pocket probing depth, and clinical attachment level. In contrast, Fischer's exact test was done to calculate bone fill. A two-tailed probability of a value less or equal to 0.05 is considered significant when comparing the mean between the groups.

Ethical Approval

Patients explained the protocols, procedure, and other details before the commencement of the study. Informed consent was obtained from the patients after presenting all the details related to the research work. The Institutional Review Board of Ahmedabad Dental College & Hospital, Ahmedabad, Gujarat 382115, India, approved this study with Reference No.: GDC/2021/237, Dated 10 June 2021. This study was planned and conducted under the declaration of Helsinki (version 2008).

RESULTS

This research was conducted to study the efficacy of subgingival lovastatin as a local drug delivery agent with its anti-inflammatory and bone-forming properties in treating patients with chronic localized moderate periodontitis. The study subjects consisted of 62 patients with chronic periodontitis. The mean age was 27.26±7.57 in the test group and 31.5±7.4 in the control group, with 12 males and 50 females.

The mean baseline values for the test and the control group for all parameters were not statistically significant (Table 1).

The gingival index did not show statistically significant results between the test and the control group at any given interval (Table 2). The mean of the modified sulcus bleeding index of the test and control groups between 0 to 15 and 0 to 90 days showed a significant variance. The change between 0 to 30 days and 0 to 60 days in the same parameter was non-significant (Table 2).

The mean pocket probing depth of the test and control groups between 0 to 15, 0 to 30, and 0 to 60 days were non-significant. The change between 0 to 90 days in the same parameter was significant (Table 2). The mean clinical attachment levels of the test and control groups between 0 to 15, 0 to 30, and 0 to 60 days were non-significant. The change between 0 to 90 days in the same parameter was significant (Table 2). Bone fills were 93.5% of the test group and 29% of the control group (Figure 2) and (Figure 3).

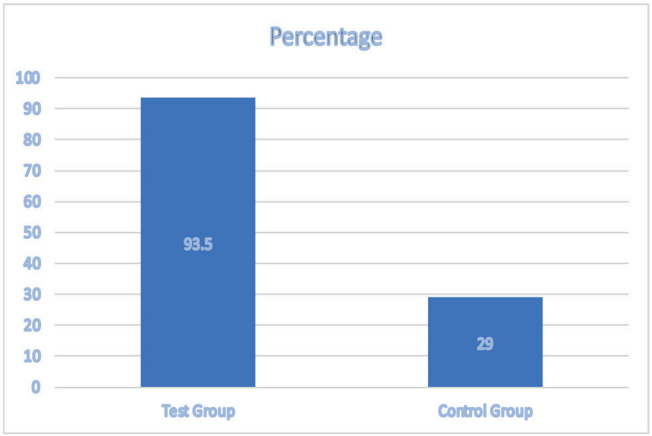

Figure 2: The Bar Chart Illustrating the Bone Fill in Both Groups.

Notes: Illustration Credit: Aayushi Gupta.

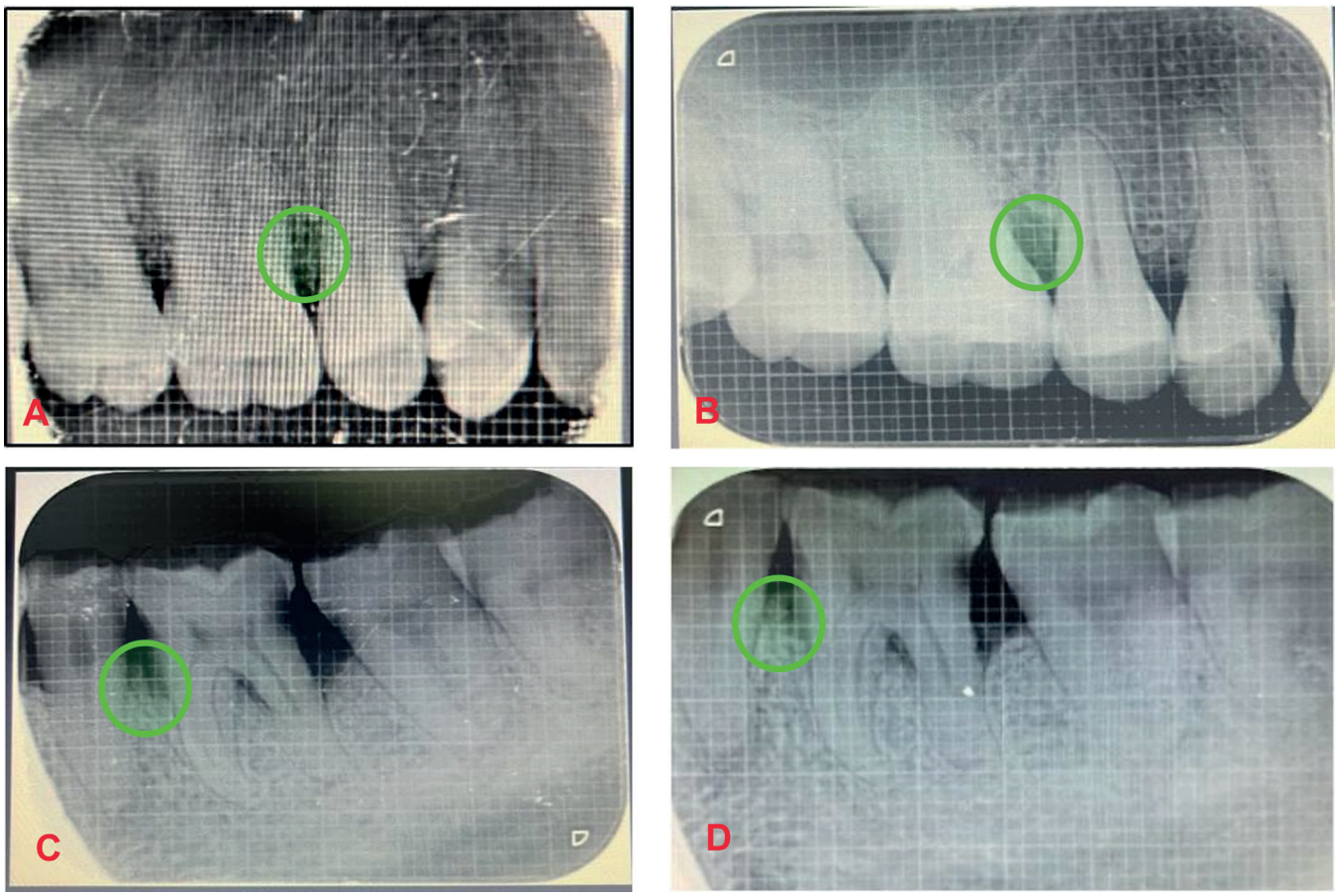

Figure 3: A: The area marked in green shows the placement of Lovastatin (Test Group). B: Bone formation is seen in the marked area (Test Group). C: The area marked in green shows the placement of the placebo gel (Control Group). D: Very Mild bone formation was seen in the marked area but was less than in the test group (Control Group).

DISCUSSION

Non-surgical therapy is the cornerstone of periodontal treatment, and local drug delivery plays a vital role [27]. The advantages of using a subgingival local drug delivery system include achieving high intrasulcular drug concentration, avoiding its systemic side effects, and better patient compliance [28]. Inflammation is the immune response to any injury [29]. In Gingival tissue, clinically, the inflammation is presented as gingivitis [30]. The progression of gingivitis to periodontitis depends on tissue hemostasis [31]. It is becoming increasingly clear that a key objective of treating inflammatory diseases is restoring tissue to its homeostasis [32].

Anti-inflammatory agents in vogue reduce inflammation and do not bring about a resolution, which is our desired goal. Therefore, a timely and appropriate host response, whether naturally occurring or induced through the supplementation of host modulatory agents, is essential to prevent the transition of an acute inflammatory response into a chronic condition and enhance the chances of restoring homeostasis. Apart from the NSAIDs, traditionally used to eliminate inflammation, a new class of lipid-lowering drugs, 3- hydroxy 2-methyl glutaryl coenzyme A reductase inhibitor, have been tried in recent years [33]. They act as competitive inhibitors of HMG-CoA reductase and are extensively used in medicine to lower cholesterol, providing an effective strategy for treating hyperlipidemia and atherosclerosis [34]. Studies have shown the effect of statins on bone regeneration when combined with calcium sulfate as a career or used alone [35,36]. Morris et al. [36] investigated the effect of injectable simvastatin on three types of periodontal defects in beagle dogs: intrabony defects, Class II furcation defects, and edentulous alveolar ridges.

Histomorphometric analysis revealed a 29% increase in ridge thickness with simvastatin. However, bone height loss and furcation defects were observed in interproximal areas. They bring about bone growth by increasing osteogenic gene expression [37]. Statins initiate restoration of tissue hemostasis through the resolution pathway as these agents can increase the production of 15-epi-lipoxins (15-epi-LxA4) and bring resolution of the acute inflammation [38]. 15-epi-lipoxins are a more bioactive form of native lipoxins, exhibiting enhanced pro-resolving properties [39]. They are generated during an acute inflammatory response by activating the lipoxin pathway, facilitated by lipid mediators of inflammation in the presence of aspirin [40]. Even though many studies show the anti-inflammatory effect of systemically taken Simvastatin and other statins, scarce data are available concerning the local delivery of statins in periodontal management [41-43]. For a drug to effect adequately, the first and foremost requirement is that it must be delivered to the site along with an appropriate career at an adequate concentration. Secondly, the drug must be able to leach out from the carrier into the surrounding milieu for a considerable period. Ideally, this drug leaching must be continued for 4 hours at a significant concentration to get the desired effect. Thus, the ideal concentration of the carrier and the drug must be developed. This clinical study aimed to find the efficacy of subgingivally delivered lovastatin as an adjunct to scaling and root planning in treating patients with chronic localized moderate periodontitis Compared to the placebo gel.

Bleeding on probing appears to be the early signs of periodontal inflammation, followed by a change in color or visual signs of inflammation [44]. Also, it is an objective sign. Non-inflamed sites rarely bleed [45]. Bleeding on probing is not a reliable indicator of progressive attachment loss but an excellent predictor of periodontal stability [46]. In the present study, the difference in modified sulcus bleeding index between 0 to 15 days was 0.50±0.81 in the test group and 0.09±0.77 in the control group with a t-value of 2.04 and p-value equal to 0.045, which was significant. On the 30th and 60th day, the mean of the test group was 0.98±0.63 and 0.66±0.64, 1.27±0.54 and 1.22±0.53, respectively, which was non-significant.

Contrary to this, on the 90th day, the difference between the test and the control group was significant (p-value 0.033). The considerable result at these intervals could be because of the anti-inflammatory properties of lovastatin gel on gingiva tissues. The reduction in bleeding on probing is consistent with the finding by Suresh et al., who stated that individuals taking locally applied statin drugs had significantly less periodontal inflammation as measured by a decrease in BOP despite equivalent plaque levels, PPD and smoking status [47]. The reduction in gingival bleeding aligns with the findings of Sakoda et al., who concluded that simvastatin exhibits an anti-inflammatory effect on human oral epithelial cells. This effect is independent of its cholesterol-lowering properties and is achieved by reducing IL-1-induced production of inflammatory cytokines, such as IL-6 and IL-8, in human oral epithelial cells [48]. The resolution of inflammation seen after statin therapy is attributed to decreased leukocyte migration [49]. Statins can suppress neutrophil transendothelial migration and chemotaxis, which helps explain their anti-inflammatory effects [50]. The anti-inflammatory effect of statins on monocytes and macrophages is attributed to their ability to reduce the expression of intercellular adhesion molecule-1 and the secretion of interleukin-6 induced by lipopolysaccharides [51]. They observed that statins reduce the Cox 2 and MMP 9 expression and their activity [52]. Statin triggered biosynthesis of the anti-inflammatory and pro-resolving mediator 15-epi-LXA4 [52].

At any interval, the gingival index showed no statistically significant change in either the test or control group. The possible reason for not getting similar results between the mSBI and GI could be due to the methods used to assess the gingival index and modified sulcus bleeding index, as the GI is used to evaluate the severity of gingival inflammation. In contrast, the mSBI assesses the severity of bleeding, a sign of gingival inflammation, and the difference in the scoring criteria.

There was also a gain in clinical attachment level in the test group on the 90th day. The mean difference in the test group was 1.32 mm and 0.87 mm in the control group. As the study subjects selected were without a gingival recession, the probing pocket depth and clinical attachment level were the same. The more significant reduction in pocket depth and clinical attachment level gain and modified sulcus bleeding index following the findings of various studies [53-55], which state that local simvastatin in the treatment of chronic periodontitis can lead to a more significant gain in CAL with decreased in the pocket depth, bleeding on probing, and gingival index.

A more excellent bone fill was observed between 0 and 90 days in the test group. This finding aligns with various studies reporting that simvastatin promotes osteoblastic activity and inhibits osteoclastic activity by stimulating bone morphogenetic protein (BMP)-induced osteoblast differentiation [56,57]. It has been shown to counteract the suppressive effects of tumor necrosis factors and prevent the inhibition of BMP-2. Additionally, it enhances alkaline phosphatase activity and mineralization while increasing the expression of bone sialoprotein, osteocalcin, and type I collagen [58]. It has been reported that statins stimulate the release of vascular endothelial growth factor (VEGF) in a dose-dependent manner and may promote osteoblast differentiation and bone nodule formation by enhancing VEGF expression in bone tissue [59].

Simvastatin significantly increased the mRNA expression of bone morphogenic protein 2 (BMP-2), vascular endothelial growth factor (VEGF), alkaline phosphatase, type I collagen, bone sialoprotein, and osteocalcin (OCN) in non-transformed osteoblastic cells (MC3T3-E1), while simultaneously suppressing the gene expression of collagenase-1 and collagenase-3 [60,61].

Limitations of the Study

The present study has limitations, including the need to conduct more research at different hospitals. Patients from different population strata should be included in the clusters. Long follow-ups are required for a more affirmative result.

CONCLUSIONS

It can be concluded from the present study that lovastatin's local drug delivery can be used as an adjunct to scaling and root planning by reducing the severity of bleeding, pocket depth reduction, and gain in clinical attachment level. It also promoted significant bone growth. The period may need longer to evaluate the long-term benefits of the desired results. Long-term studies with larger sample sizes are required to assess the benefit of lovastatin local drug delivery, its release pattern, minimum inhibitory concentration, whether multiple doses are needed, and its effect on inflammation.

Consent for Publication

The author reviewed and approved the final version and has agreed to be accountable for all aspects of the work, including any accuracy or integrity issues.

DISCLOSURE

The author declares that they do not have any financial involvement or affiliations with any organization, association, or entity directly or indirectly related to the subject matter or materials presented in this review paper. This includes honoraria, expert testimony, employment, ownership of stocks or options, patents, or grants received or pending royalties.

Data Availability

Information for this original paper is available only for research purpose only.

Authorship Contribution

All authors contributed significantly to the work, whether in the conception, design, utilization, collection, analysis, and interpretation of data or all these areas. They also participated in the paper's drafting, revision, or critical review, gave their final approval for the version that would be published, decided on the journal to which the article would be submitted, and made the responsible decision to be held accountable for all aspects of the work.

REFERENCES

1. Paul O, Arora P, Mayer M, Chatterjee S. Inflammation in Periodontal Disease: Possible Link to Vascular Disease. *Front Physiol*. 2021; **11**: 609614. 10.3389/fphys.2020.609614
2. Kumar PS. Microbial dysbiosis: The root cause of periodontal disease. *J Periodontol*. 2021; **92**(8):1079-1087. doi: 10.1002/JPER.21-0245.
3. Sedghi LM, Bacino M, Kapila YL. Periodontal Disease: The Good, The Bad, and The Unknown. *Front Cell Infect Microbiol*. 2021;**11**:766944. doi: 10.3389/fcimb.2021.766944.
4. Loesche WJ. The antimicrobial treatment of periodontal disease: changing the treatment paradigm. *Crit Rev Oral Biol Med*. 1999;**10**(3):245-75. doi: 10.1177/10454411990100030101.
5. Rosier BT, De Jager M, Zaura E, Krom BP. Historical and contemporary hypotheses on the development of oral diseases: are we there yet? *Front Cell Infect Microbiol*. 2014; **4**:92. doi: 10.3389/fcimb.2014.00092
6. Brookes ZLS, Bescos R, Belfield LA, Ali K, Roberts A. Current uses of chlorhexidine for management of oral disease: a narrative review. *J Dent*. 2020; **103**:103497. doi: 10.1016/j.jdent.2020.103497
7. Sharifipour E, Shams S, Esmkhani M, Khodadadi J, Fotouhi-Ardakani R, Koohpaei A, Doosti Z, Ej Golzari S. Evaluation of bacterial co-infections of the respiratory tract in COVID-19 patients admitted to ICU. *BMC Infect Dis*. 2020;**20**(1):646. doi: 10.1186/s12879-020-05374-z.
8. Alassy H, Pizarek JA, Kormas I, Pedercini A, Wolff LF. Antimicrobial adjuncts in the management of periodontal and peri-implant diseases and conditions: A narrative review. *Front Oral and Maxillofac Med*. 2021; **3**:1-19. doi: 10.21037/fomm-20-84
9. Toledano-Osorio M, Vallecillo C, Toledano R, Aguilera FS, Osorio MT, Muñoz-Soto E, García-Godoy F, Vallecillo-Rivas M. A Systematic Review and Meta-Analysis of Systemic Antibiotic Therapy in the Treatment of Peri-Implantitis. *Int J Environ Res Public Health*. 2022;**19**(11):6502. doi: 10.3390/ijerph19116502.
10. Ezike TC, Okpala US, Onoja UL, Nwike CP, Ezeako EC, Okpara OJ, Okoroafor CC, Eze SC, Kalu OL, Odoh EC, Nwadike UG, Ogbodo JO, Umeh BU, Ossai EC, Nwanguma BC. Advances in drug delivery systems, challenges, and future directions. *Heliyon*. 2023; **9**(6):e17488. doi: 10.1016/j.heliyon.2023.e17488.
11. Budală DG, Luchian I, Tatarciuc M, Butnaru O, Armencia AO, Virvescu DI, Scutariu MM, Rusu D. Are Local Drug Delivery Systems a Challenge in Clinical Periodontology? *J Clin Med*. 2023; **12**(12):4137. doi: 10.3390/jcm12124137.
12. Vigani B, Rossi S, Sandri G, Bonferoni MC, Caramella CM, Ferrari F. Recent Advances in the Development of In Situ Gelling Drug Delivery Systems for Non-Parenteral Administration Routes. *Pharmaceutics*. 2020; **12**:859. doi: 10.3390/pharmaceutics12090859
13. Lengyel M, Kállai-Szabó N, Antal V, Laki AJ, Antal I. Microparticles, Microspheres, and Microcapsules for Advanced Drug Delivery. *Sci Pharm.* 2019; **87**:(3). doi:10.3390/scipharm87030020.
14. Mashabela LT, Maboa MM, Miya NF, Ajayi TO, Chasara RS, Milne M, Mokhele S, Demana PH, Witika BA, Siwe-Noundou X, Poka MS. A Comprehensive Review of Cross-Linked Gels as Vehicles for Drug Delivery to Treat Central Nervous System Disorders. *Gels*. 2022; **8** (9):563. doi: 10.3390/gels8090563.
15. Puri K, Puri N. Local drug delivery agents as adjuncts to endodontic and periodontal therapy. *J Med Life*. 2013; **6**:414-9. https://pmc.ncbi.nlm.nih.gov/articles/PMC4034307/pdf/JMedLife-06-414.pdf
16. Bessa LJ, Botelho J, Machado V, Alves R, Mendes JJ. Managing Oral Health in the Context of Antimicrobial Resistance. *Int J Environ Res Public Health*. 2022; **19**:16448. doi: 10.3390/ijerph192416448
17. Caruana EJ, Roman M, Hernández-Sánchez J, Solli P. Longitudinal studies. *J Thorac Dis*. 2015; **7**(11): E537-40. doi: 10.3978/j.issn.2072-1439.2015.10.63.
18. BioRender. (2024). Available at: https://BioRender.com/f35s380 Accessed: December 6, 2024.
19. Löe H. The Gingival Index, the Plaque Index, and the Retention Index Systems. *J Periodontol*. 1967;**38**(6):Suppl:610-6. doi: 10.1902/jop.1967.38.6.610.
20. Newbrun E. Indices to Measure Gingival Bleeding. *J Periodont*. 1996; **67**:555-61. doi: 10.1902/jop.1996.67.6.555.
21. Tobias G, Spanier AB. Modified Gingival Index (MGI) Classification Using Dental Selfies. *App Sci*. 2020; **10**(24):8923. https://doi.org/10.3390/app10248923
22. Chung HM, Park JY, Ko KA, Kim CS, Choi SH, Lee JS. Periodontal probing on digital images compared to clinical measurements in periodontitis patients. *Sci Rep*. 2022;**12**(1):1616. doi:10.3390/app10248923.
23. Mishra P, Bhargava A, Nigam-Gupta N. A pilot study to evaluate the effectiveness of adjunctive use of two antimicrobial topical gels in chronic gingivitis. *J Clin Exp Dent*. 2021;**13**(4):e342-e349. doi: 10.4317/jced.57635.
24. Farook FF, Alodwene H, Alharbi R, Alyami M, Alshahrani A, Almohammadi D, Alnasyan B, Aboelmaaty W. Reliability assessment between clinical attachment loss and alveolar bone level in dental radiographs. *Clin Exp Dent Res*. 2020;**6**(6):596-601. doi: 10.1002/cre2.324.

25. Siddique SN, Anwar MA, Zaman H, Haider I, Ahmad A, Umair M, Baig MA. Quality Assessment of Periapical Radiographs Taken by Dental Assistants Using the Recent Faculty of General Dental Practice (FGDP) Guidelines. *Cureus*. 2024;**16**(9):e68508. doi: 10.7759/cureus.68508.
26. Adurty C, Tejaswi KS, Shivani CRN, Navya D, Gopinath C, Dhulipalla R. Accuracy of digital intraoral periapical radiography and cone-beam computed tomography in the measurement of intrabony defects: A comparative study. *J Indian Soc Periodontol*. 2021; **25** (6):491-495. doi: 10.4103/jisp.jisp_518_20.
27. Liang J, Peng X, Zhou X, Zou J, Cheng L. Emerging Applications of Drug Delivery Systems in Oral Infectious Diseases Prevention and Treatment. *Molecules*. 2020; **25**:516. doi: 10.3390/molecules25030516
28. Sholapurkar A, Sharma D, Glass B, Miller C, Nimmo A, Jennings E. Professionally Delivered Local Antimicrobials in the Treatment of Patients with Periodontitis—A Narrative Review. *Dent J (Basel)*. 2020; **9**:2. doi: 10.3390/dj9010002
29. Muire PJ, Mangum LH, Wenke JC. Time Course of Immune Response and Immunomodulation During Normal and Delayed Healing of Musculoskeletal Wounds. *Front Immunol*. 2020; **11**:1056. doi: 10.3389/fimmu.2020.01056
30. Bamashmous S, Kotsakis GA, Kerns KA, Leroux BG, Zenobia C, Chen D, Trivedi HM, McLean JS, Darveau RP. Human variation in gingival inflammation. *Proc Natl Acad Sci USA*. 2021; **118**(27):e2012578118. doi: 10.1073/pnas.2012578118.
31. Könönen E, Gursoy M, Gursoy UK. Periodontitis: A Multifaceted Disease of Tooth-Supporting Tissues. *J Clin Med*. 2019; **8**:1135. 10.3390/jcm8081135
32. Schett G, Neurath MF. Resolution of chronic inflammatory disease: universal and tissue-specific concepts. *Nat Commun*. 2018; **9**:3261. doi: 10.1038/s41467-018-05800-6
33. Morofuji Y, Nakagawa S, Ujifuku K, Fujimoto T, Otsuka K, Niwa M, Tsutsumi K. Beyond Lipid-Lowering: Effects of Statins on Cardiovascular and Cerebrovascular Diseases and Cancer. *Pharmaceuticals (Basel)*. 2022;**15**(2):151. doi: 10.3390/ph15020151.
34. Markowska A, Antoszczak M, Markowska J, Huczyński A. Statins: HMG-CoA Reductase Inhibitors as Potential Anticancer Agents against Malignant Neoplasms in Women. *Pharmaceuticals*. 2020; **13**:422. 10.3390/ph13120422
35. Cruz R, Pesce G, Calasans-Maia J, Moraschini V, Calasans-Maia MD, Granjeiro JM. Calcium Phosphate Carrying Simvastatin Enhances Bone Regeneration: A Systematic Review. *Braz Dent J*. 2020; **31**:93-102. doi: 10.1590/0103-6440202002971
36. Venkatesan N, Liyanage ADT, Castro-Núñez J, Asafo-Adjei T, Cunningham LL, Dziubla TD, Puleo DA. Biodegradable Polymerized Simvastatin Stimulates Bone Formation. *Acta Biomater*. 2019; **93**:192-9. doi: 10.1016/j.actbio.2019.04.059
37. Morris MS, Lee Y, Lavin MT, Giannini PJ, Schmid MJ, Marx DB, Reinhardt RA. Injectable simvastatin in periodontal defects and alveolar ridges: pilot studies. *J Periodontol*. 2008; **79**:1465-73. 10.1902/jop.2008.070659
38. Undas A. Statins in preventing thromboembolic events: from seminal studies to recent advances. *Pol Arch Intern Med*. 2022; **132**:1-8. doi: 10.20452/pamw.16208
39. Jaén RI, Sánchez-García S, Fernández-Velasco M, Boscá L, Prieto P. Resolution-Based Therapies: The Potential of Lipoxins to Treat Human Diseases. *Front Immunol*. 2021;**12**:658840. doi: 10.3389/fimmu.2021.658840.
40. Kain V, Liu F, Kozlovskaya V, Ingle KA, Bolisetty S, Agarwal A, Khedkar S, Prabhu SD, Kharlampieva E, Halade GV. Resolution Agonist 15-epi-Lipoxin A4 Programs Early Activation of Resolving Phase in Post-Myocardial Infarction Healing. *Sci Rep*. 2017;**7**(1):9999. doi: 10.1038/s41598-017-10441-8.
41. Proute MC, Kothur N, Georgiou P, Serhiyenia T, Shi W, Kerolos ME, Pradeep R, Akram A, Khan S. The Effect of Statin Therapy on Inflammatory Biomarkers: A Systematic Review. *Cureus*. 2021; **13**(9):e18273. doi: 10.7759/cureus.18273.
42. Kim SW, Kang HJ, Jhon M, Kim JW, Lee JY, Walker AJ, Agustini B, Kim JM, Berk M. Statins and Inflammation: New Therapeutic Opportunities in Psychiatry. *Front Psychiat*. 2019;**10**:103. doi: 10.3389/fpsyt.2019.00103.
43. Li G-M, Zhao J, Li B, Zhang X-F, Ma J-X, Ma X-L, Liu J. The anti-inflammatory effects of statins on patients with rheumatoid arthritis: A systemic review and meta-analysis of 15 randomized controlled trials. *Autoimmun Rev*. 2018; **17**:215-25. doi: 10.1016/j.autrev.2017.10.013
44. Ali Alftaikhah SA, Issrani R, Alnasser M, Almutairi HA, Khattak O, Iqbal A, Prabhu N. Salivary Biomarkers in Periodontitis: A Scoping Review. *Cureus*. 2023; **15**(12):e50207. doi: 10.7759/cureus.50207.
45. Rathee M, Jain P. Gingivitis. [Updated 2023 Mar 27]. In: StatPearls [Internet]. Treasure Island (FL): StatPearls Publishing; 2024. Available at https://www.ncbi.nlm.nih.gov/books/NBK557422/ [Accessed December 21, 2024]
46. Dukka H, Saleh MHA, Ravidà A, Greenwell H, Wang HL. Is bleeding on probing a reliable clinical indicator of peri-implant diseases? *J Periodontol*. 2021; **92**:1669-1674. doi:10.1002/JPER.20-0890
47. Suresh S, Narayana S, Jayakumar P, Sudhakar U, Pramod V. Evaluation of anti-inflammatory effect of statins in chronic periodontitis. *Indian J Pharmacol*. 2013; **45**:391-4.

doi:10.4103/0253-7613.115017

48. Sakoda K, Yamamoto M, Negishi Y, Liao JK, Node K, Izumi Y. Simvastatin decreases IL-6 and IL-8 production in epithelial cells. *J Dent Res*. 2006; **85**:520-3. doi:10.1177/154405910608500608
49. Alfaro S, Acuña V, Ceriani R, Cavieres MF, Weinstein-Oppenheimer CR, Campos-Estrada C. Involvement of Inflammation and Its Resolution in Disease and Therapeutics. *Int J Mol Sci*. 2022; **23**:10719. doi:10.3390/ijms231810719
50. Lin W-C, Fessler MB. Regulatory mechanisms of neutrophil migration from the circulation to the airspace. *Cell Mol Life Sci*. 2021; **78**:4095-124. doi:10.1007/s00018-021-03768-z
51. Koushki K, Shahbaz SK, Mashayekhi K, Sadeghi M, Zayeri ZD, Taba MY, Banach M, Al-Rasadi K, Johnston TP, Sahebkar A. Anti-inflammatory Action of Statins in Cardiovascular Disease: The Role of Inflammasome and Toll-Like Receptor Pathways. *Clin Rev Allergy Immunol*. 2021; **60**(2):175-199. doi: 10.1007/s12016-020-08791-9.
52. Marcianò G, Palleria C, Casarella A, Rania V, Basile E, Catarisano L, Vocca C, Bianco L, Pelaia C, Cione E, D'Agostino B, Citraro R, De Sarro G, Gallelli L. Effect of Statins on Lung Cancer Molecular Pathways: A Possible Therapeutic Role. *Pharmaceuticals (Basel)*. 2022; **15**(5):589. doi: 10.3390/ph15050589.
53. Pradeep AR, Thorat MS. Clinical effect of subgingivally delivered simvastatin in the treatment of patients with chronic periodontitis: a randomized clinical trial. *J Periodontol*. 2010; **81**:214-22. doi: 10.1902/jop.2009.090429
54. Agarwal S, Chaubey KK, Chaubey A, Agarwal V, Madan E, Agarwal MC. Clinical efficacy of subgingivally delivered simvastatin gel in chronic periodontitis patients. *J Indian Soc Periodontol*. 2016; **20**:409-16. 10.4103/0972-124X.194270
55. Cao R, Li Q, Chen Y, Yao M, Wu Q, Zhou H. Efficacy of locally-delivered statins adjunct to non-surgical periodontal therapy for chronic periodontitis: a Bayesian network analysis. *BMC Oral Health*. 2019; **19**:105. 10.1186/s12903-019-0789-2
56. S Martande S, Kumari M, Pradeep AR, Pal Singh S, Kumar Suke D. Comparative evaluation of the efficacy of subgingivally delivered 1.2% Atorvastatin and 1.2% Simvastatin in the treatment of intrabony defects in chronic periodontitis: a randomized controlled trial. *J Dent Res Dent Clin Dent Prospects*. 2017;**11**(1):18-25. doi: 10.15171/joddd.2017.004.
57. Xu R, Shi G, Xu L, Gu Q, Fu Y, Zhang P, Cheng J, Jiang H. Simvastatin improves oral implant osseointegration via enhanced autophagy and osteogenesis of BMSCs and inhibited osteoclast activity. *J Tissue Eng Regen Med*. 2018;**12**(5):1209-1219. doi: 10.1002/term.2652.
58. Kitaura H, Marahleh A, Ohori F, Noguchi T, Nara Y, Pramusita A, Kinjo R, Ma J, Kanou K, Mizoguchi I. Role of the Interaction of Tumor Necrosis Factor-α and Tumor Necrosis Factor Receptors 1 and 2 in Bone-Related Cells. *Int J Mol Sci*. 2022;**23**(3):1481. doi: 10.3390/ijms23031481.
59. Granat MM, Eifler-Zydel J, Kolmas J. Statins-Their Role in Bone Tissue Metabolism and Local Applications with Different Carriers. *Int J Mol Sci*. 2024;**25**(4):2378. doi: 10.3390/ijms25042378.
60. Chen P-Y, Sun J-S, Tsuang Y-H, Chen M-H, Weng P-W, Lin F-H. Simvastatin promotes osteoblast viability and differentiation via Ras/Smad/Erk/BMP-2 signaling pathway. *Nutr Res*. 2010; **30**:191-9. 10.1016/j.nutres.2010.03.004
61. Park JB, Zhang H, Lin CY, Chung CP, Byun Y, Park YS, Yang VC. Simvastatin maintains osteoblastic viability while promoting differentiation by partially regulating the expressions of estrogen receptors α. *J Surg Res*. 2012;**174**(2):278-83. doi: 10.1016/j.jss.2010.12.029.