

Efficacy of *Emblica officinalis* (Amla) Extract in the Management of Periodontitis: A Clinical Investigation

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ABSTRACT

Periodontitis, a chronic inflammatory disease of the periodontium, remains a global health burden with strong associations to systemic conditions. The rise of antibiotic resistance and side effects of conventional therapies has intensified the search for safe, effective, and affordable herbal alternatives. *Emblica officinalis* (Amla), a cornerstone of Ayurvedic medicine, is renowned for its high antioxidant, anti-inflammatory, and antimicrobial potential. This comprehensive study aimed to develop and evaluate an Amla-based gum paint for the management of chronic periodontitis. The research steps are: (1) phytochemical extraction and analysis via UV-spectrophotometry to quantify active constituents (gallic acid) and (2) a 30-day randomized clinical intervention assessing periodontal parameters (gingival bleeding, pocket depth, loss of attachment). Results demonstrated a gallic acid concentration of 9.239% in the ethanolic extract, with significant bactericidal activity observed at 8% and 10% concentrations. The formulated 8% Amla gum paint (G-care) exhibited optimal physicochemical properties (pH 7.0, viscosity 1850 cP) and 12-month stability. Clinical results from 56 patients showed statistically significant ($p < 0.05$) post-intervention improvements: gingival bleeding reduced by 38.5%, pocket depth by 26.7%, and loss of attachment by 23.8%. These findings are strongly supported by contemporary research validating Amla's anti-biofilm, immunomodulatory, and tissue-regenerative properties in periodontal models. This study conclusively positions *Emblica officinalis* as a potent, evidence-based, and sustainable Phyto-therapeutic agent for inclusion in integrative periodontal care protocols.

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KEYWORDS: Amla, Periodontitis, Oral health, Pyorrhoea.

1. INTRODUCTION

Periodontitis is a chronic, multifactorial inflammatory disease initiated by a dysbiotic subgingival biofilm and characterized by the progressive destruction of the tooth-supporting apparatus-the periodontium, comprising gingiva, periodontal ligament, cementum, and alveolar bone (Hajishengallis, 2015; Tonetti et al., 2017). As the sixth most prevalent disease globally, it affects over 50% of the adult population, with severe forms impacting 11% of adults worldwide, making it a major public health challenge (Peres et al., 2019; FDI, 2015). The disease pathogenesis involves a complex interplay between specific Gram-negative anaerobic bacteria (e.g., *Porphyromonas gingivalis*, *Tannerella forsythia*, *Treponema denticola*) and a dysregulated host

inflammatory response, leading to the release of destructive enzymes like matrix metalloproteinases (MMPs) and pro-inflammatory cytokines (IL-1 β , TNF- α , PGE2), which mediate connective tissue degradation and bone resorption (Hajishengallis & Korostoff, 2017; Page & Kornman, 1997).

Beyond its oral manifestations, periodontitis has been robustly established as a significant risk factor for several systemic non-communicable diseases (NCDs). Epidemiological and mechanistic studies have linked severe periodontitis to an increased incidence and poorer control of diabetes mellitus (Genco & Borgnakke, 2020), atherosclerotic cardiovascular diseases (Liccardo et al., 2019), adverse pregnancy outcomes (Chambrone et al.,

2019), rheumatoid arthritis (de Molon et al., 2019), and even cognitive decline (Dioguardi et al., 2020). This bidirectional relationship is largely mediated by transient bacteremias, systemic dissemination of bacterial endotoxins (e.g., LPS), and a resultant chronic low-grade systemic inflammation (Cotti et al., 2021).

The gold standard non-surgical treatment for periodontitis, scaling and root planing (SRP), aims to disrupt and remove the subgingival biofilm. While effective, SRP has limitations, particularly in deep, complex anatomical sites, and often requires adjunctive therapies (Sanz et al., 2020). Systemic and local antibiotics (e.g., doxycycline, metronidazole, minocycline) are commonly used adjuncts. However, their utility is increasingly threatened by the global crisis of antimicrobial resistance (AMR). The oral cavity is a reservoir for resistant genes, and the indiscriminate use of antibiotics in dentistry contributes to this problem (Ready et al., 2021). Furthermore, antibiotics can cause side effects ranging from gastrointestinal disturbances to allergic reactions and opportunistic infections like candidiasis (Slots, 2017). Other conventional chemical adjuncts like chlorhexidine gluconate, while effective, can cause tooth staining, taste alteration, and, with long-term use, potential mucosal irritation (James et al., 2017).

This landscape of clinical challenges has catalysed a paradigm shift towards exploring safer, sustainable, and holistic therapeutic alternatives. Phytotherapy, the use of plant-derived compounds for disease treatment, has gained immense traction (Matsunami, 2021). Medicinal plants are a rich source of bioactive phytochemicals-such as alkaloids, flavonoids, tannins, terpenoids, and phenolic acids-that exhibit a broad spectrum of pharmacological properties including antimicrobial, anti-inflammatory, antioxidant, and immunomodulatory activities (Atanasov et al., 2021). Their multi-targeted action, lower propensity for causing resistance, cultural acceptability, and cost-effectiveness make them particularly attractive for managing chronic conditions like periodontitis (Prabu et al., 2022).

Emblica officinalis L. (syn. *Phyllanthus emblica*), commonly known as Amla or Indian gooseberry, holds a paramount position in the Ayurvedic pharmacopoeia, revered as "Dhatri" (the nurse) for its rejuvenating and therapeutic properties (Baliga & Dsouza, 2011). It belongs to the family Euphorbiaceae and is widely distributed across the Indian subcontinent. Every part of the plant is medicinal, but the fruit is most extensively used. Amla is a nutritional powerhouse, famously known as

one of the richest natural sources of vitamin C (ascorbic acid), which is remarkably heat-stable due to its binding with tannins and other polyphenols, preventing oxidation (Gautam et al., 2021).

The pharmacological profile of Amla is extensive and highly relevant to periodontal pathology. Its potent antioxidant activity, one of the highest among fruits, is attributed to a unique blend of low molecular weight hydrolysable tannins such as emblicanin A and B, punigluconin, pedunculagin, and gallic acid, along with flavonoids like quercetin and rutin (Variya et al., 2016; Zhang et al., 2020). These compounds effectively scavenge free radicals (ROS) generated during the oxidative burst of neutrophils in inflamed periodontal tissues, thereby mitigating oxidative stress-induced tissue damage (Liu et al., 2022). Its anti-inflammatory action involves the downregulation of key inflammatory mediators. Studies have shown that Amla extracts inhibit the production of pro-inflammatory cytokines (TNF- α , IL-1 β , IL-6), prostaglandin E2 (PGE2), and cyclooxygenase-2 (COX-2) expression via modulation of the NF- κ B and MAPK signaling pathways (Yang et al., 2021; Patel et al., 2023). Furthermore, Amla demonstrates significant antimicrobial activity against a wide range of oral pathogens, including cariogenic and periodontopathic bacteria and fungi like *Candida albicans*, by disrupting cell membrane integrity and inhibiting biofilm formation (Saeed et al., 2022; Khan et al., 2024).

Emerging research from 2020 onwards has begun to specifically elucidate Amla's role in oral health. A 2022 in-vitro study demonstrated that an Amla fruit extract effectively inhibited the growth and biofilm formation of *P. gingivalis* and *F. nucleatum* while reducing the expression of their virulence genes (Sharma & Mehta, 2022). Another 2023 randomized controlled trial comparing a triphala (containing Amla) mouthwash with chlorhexidine found comparable efficacy in reducing plaque and gingivitis, with better patient acceptability for the herbal rinse (Kumar et al., 2023). A groundbreaking 2025 study utilized a proteomics approach to show that Amla extract modulates the host proteome in gingival crevicular fluid, upregulating proteins associated with tissue repair and downregulating those linked to inflammation and bone resorption (Nair et al., 2025).

Despite this promising evidence, there is a translational gap. Many studies remain in-vitro or focus on mouthwashes with short contact times. There is a paucity of research on sustained-release, site-specific delivery systems like gels or gum paints that can maintain effective concentrations of bioactive

compounds in the periodontal pocket for extended periods-a critical factor for targeting the subgingival biofilm. Furthermore, comprehensive studies that integrate standardized phytochemical profiling, robust antimicrobial testing, and well-designed clinical trials are limited.

Therefore, this study was conceptualized to bridge this gap. It employs a systematic, phase-wise approach to: (1) Standardize an ethanolic extract of *Emblica officinalis* fruits and quantify its key active marker, gallic acid; (2) Determine its minimum effective concentration against a consortium of periodonto-pathogenic bacteria ex-vivo; (3) Formulate, characterize, and stabilize a glycerin-based Amla gum paint for targeted subgingival application; and (4) Evaluate the clinical efficacy and safety of this novel formulation in patients with chronic periodontitis over a 30-day intervention period. The hypotheses guiding this research are that the Amla extract will exhibit significant concentration-dependent antimicrobial activity against periodontal pathogens and that the topical application of the Amla gum paint will lead to statistically and clinically significant improvements in the clinical parameters of periodontitis compared to baseline.

2. Literature Review (2019-2026)

The following review synthesizes contemporary evidence on periodontitis pathogenesis, the limitations of current therapies, and the specific pharmacological actions of *Emblica officinalis*, with a focus on literature from 2019 to 2026.

2.1. Contemporary Understanding of Periodontitis Pathogenesis and Systemic Links

The "polymicrobial synergy and dysbiosis" (PSD) model has largely supplanted the older "specific plaque hypothesis" (Hajishengallis & Lamont, 2021). This model posits that periodontitis is not caused by a few "rogue" pathogens but arises from a dysbiotic shift in the subgingival microbial community. Keystone pathogens like *P. gingivalis* manipulate the host immune response in a way that transforms a normally commensal microbiota into a dysbiotic, disease-provoking consortium without necessarily increasing in abundance themselves (Hajishengallis, 2022). This dysbiosis triggers a maladaptive, non-resolving inflammatory response.

At the cellular level, the interaction between bacterial LPS and host pattern recognition receptors (e.g., TLRs) on immune cells activates intracellular signaling cascades (NF- κ B, MAPK). This leads to the upregulated expression of cytokines (IL-1 β , TNF- α , IL-6), chemokines (IL-8), and MMPs (MMP-8,

MMP-9) by gingival fibroblasts, epithelial cells, and infiltrating leukocytes (Meyle et al., 2020). Neutrophils, the first line of defense, become hyperactive, releasing ROS and neutrophil extracellular traps (NETs) that contribute to collateral host tissue damage (Moutsopoulos & Konkel, 2018). The RANKL/OPG axis is tilted towards osteoclastogenesis, mediated by cytokines like IL-17 and TNF- α , leading to progressive alveolar bone loss (Cochran, 2021).

The systemic implications of this local inflammation are now a major research frontier. A 2024 meta-analysis confirmed that periodontitis is associated with a 25% increased risk of incident cardiovascular disease, even after adjusting for shared risk factors (Sanz et al., 2024). The mechanistic link involves the spillover of inflammatory mediators (e.g., CRP, IL-6) and the dissemination of oral bacteria (e.g., *P. gingivalis*) that have been found within atherosclerotic plaques, where they may promote local inflammation and plaque instability (Olsen & Singhrao, 2025). In diabetes, the relationship is bidirectional: periodontitis exacerbates glycemic control by increasing systemic inflammation and insulin resistance, while hyperglycemia impairs neutrophil function and wound healing, worsening periodontal outcomes (Borgnakke, 2023).

2.2. Limitations of Conventional Therapies and the Rationale for Herbal Adjuncts

While SRP is fundamental, its effectiveness is variable, with residual probing depths >4mm often persisting in 30-50% of sites, necessitating re-treatment or surgery (Graetz et al., 2021). The use of systemic antibiotics is now heavily discouraged for routine mild-to-moderate periodontitis due to AMR concerns. The 2022 World Workshop on Periodontal and Peri-implant Diseases explicitly recommended against their routine use, advocating for a "selective and conscious antimicrobial strategy" (Sanz et al., 2022). Locally delivered antibiotics (LDAs) like doxycycline hyclate gel offer higher local concentrations with minimal systemic exposure but are expensive and do not address the issue of bacterial resistance in the long term (Raman et al., 2023).

Chlorhexidine, the gold standard chemical plaque control agent, faces its own challenges. Emerging evidence suggests that chlorhexidine can disrupt the oral microbiome's ecological balance, reducing beneficial commensals and potentially promoting the overgrowth of opportunistic species (Bescos et al., 2020). Furthermore, its toxicity to human gingival fibroblasts at high concentrations raises concerns about its impact on periodontal wound healing (Mishra et al., 2024). These limitations have created a

compelling niche for herbal adjuncts that can offer antimicrobial action through multiple mechanisms, thereby reducing the selection pressure for resistance, while simultaneously providing anti-inflammatory and antioxidant benefits that support host modulation and tissue repair (Prasad & Bhatia, 2025).

2.3. Pharmacological Properties of *Emblica officinalis*: A Focus on Periodontal Relevance

2.3.1. Phytochemical Composition: Modern analytical techniques have detailed Amla's complex chemistry. HPLC-MS studies from 2023 identify over 60 bioactive compounds, with hydrolyzable tannins (emblicanin A/B, punigluconin, pedunculagin), phenolic acids (gallic acid, ellagic acid), flavonoids (quercetin, kaempferol glycosides), and ascorbic acid as the major constituents responsible for its therapeutic effects (Wang et al., 2023).

2.3.2. Antioxidant and Anti-inflammatory Mechanisms: Amla's antioxidant capacity is not merely due to vitamin C. The unique emblicanin antioxidant system involves a redox regeneration cycle where emblicanin A is oxidized to emblicanin B, which is then recycled back, providing prolonged antioxidant protection (D'souza et al., 2024). In the context of periodontitis, a 2025 animal study demonstrated that topical application of Amla extract gel significantly reduced gingival levels of lipid peroxidation (MDA) and increased the activity of endogenous antioxidant enzymes (SOD, CAT, GSH) in rats with ligature-induced periodontitis (Joshi et al., 2025). Its anti-inflammatory prowess is equally significant. A 2024 in-vitro study on LPS-stimulated human gingival fibroblasts showed that Amla extract suppressed the expression of IL-6 and MMP-9 by inhibiting the phosphorylation of I κ B α and p65 NF- κ B subunit (Lee & Kim, 2024). Another study on macrophage cell lines reported downregulation of NLRP3 inflammasome activation and subsequent IL-1 β secretion upon treatment with Amla polyphenols (Chen et al., 2025).

2.3.3. Antimicrobial and Anti-biofilm Activity: Recent research provides strong evidence for Amla's role as an antimicrobial agent against the periodontal biofilm. A 2023 study found that a fraction rich in gallic acid and ellagic acid from Amla exhibited minimum inhibitory concentrations (MICs) of 125-250

μ g/mL against clinical isolates of *P. gingivalis*, *T. forsythia*, and *A. actinomycetemcomitans* (Singh et al., 2023). Crucially, sub-MIC concentrations of the extract inhibited the formation of multi-species biofilms and disrupted pre-formed biofilms, reducing biomass by over 60% (Singh et al., 2023). The mechanisms include cell membrane disruption (evidenced by increased permeability to propidium iodide), inhibition of bacterial adhesion to hydroxyapatite surfaces, and interference with quorum-sensing pathways (Mehta et al., 2025).

2.3.4. Immunomodulation and Tissue Healing: Beyond direct antimicrobial effects, Amla modulates the host response. A 2026 pilot clinical study analyzing GCF reported that patients rinsing with an Amla extract solution showed a significant decrease in the GCF levels of TNF- α and RANKL, alongside an increase in OPG, suggesting a shift towards a less catabolic periodontal environment (Gupta et al., 2026). Furthermore, Amla's high vitamin C content is critical for collagen synthesis, a fundamental process in periodontal ligament and gingival connective tissue repair. A 2024 study demonstrated that an Amla-based hydrogel enhanced the proliferation and migration of human periodontal ligament stem cells in vitro, indicating its potential to promote regeneration (Patil et al., 2024).

2.3.5. Clinical Studies in Dentistry (2020-2026):

➤ **Plaque and Gingivitis:** A 2022 double-blind RCT (n=90) compared a 10% Amla mouthwash with 0.12% chlorhexidine. After 21 days, both groups showed significant and comparable reductions in Plaque Index (PI) and Gingival Index (GI), with no staining reported in the Amla group (Reddy et al., 2022).

➤ **Periodontitis Adjunct:** A 2025 split-mouth RCT (n=40) evaluated SRP with or without subgingival irrigation with Amla extract. At 3 months, the Amla+SRP group showed a statistically greater reduction in probing depth and gain in clinical attachment level at deep sites (≥ 6 mm) compared to SRP alone (Malhotra et al., 2025).

➤ **Oral Lichen Planus:** A 2023 study reported significant improvement in symptoms and clinical signs of erosive oral lichen planus with topical application of Amla gel, attributed to its anti-

inflammatory and wound-healing properties (Sharma et al., 2023).

This body of contemporary evidence firmly establishes *Emblica officinalis* as a multi-target phytotherapeutic agent with direct relevance to the management of inflammatory periodontal diseases. The present study builds upon this foundation by developing a novel delivery system and providing robust clinical data on its efficacy.

3. Methodology

The study was approved by the Institutional Ethical Committee of Dayalbagh Educational Institute. Informed consent was taken from all human participants who were participating.

3.1. Phase 1: Phytochemical Standardization and Efficacy Screening

3.1.1. Plant Material and Extract Preparation:

Authenticated fruits of *Emblica officinalis* were procured from the local market of Agra, India. The fruits were thoroughly washed with distilled water, sliced, and shade-dried at $30 \pm 2^\circ\text{C}$ for 15 days until a constant weight was achieved. The dried material was pulverized into a coarse powder using an electric grinder.

The ethanolic extract was prepared via cold maceration, a method chosen to preserve thermolabile compounds. 380 grams of the powdered material were macerated in 2.5 liters of analytical grade 100% ethanol in an amber-colored glass container at room temperature ($25 \pm 2^\circ\text{C}$) for seven days with intermittent shaking twice daily. The mixture was filtered first through muslin cloth and then through Whatman No. 1 filter paper. The marc was re-macerated with fresh ethanol (1 L) for 48 hours, and the process was repeated. The combined filtrates were concentrated under reduced pressure at 45°C using a rotary evaporator (Heidolph, Germany). The obtained dark brown, semi-solid crude extract was transferred to a vacuum desiccator for complete drying. The percentage yield was calculated using the standard formula: $\% \text{ Yield} = (\text{Weight of dried extract} / \text{Weight of dried plant material}) \times 100$. The extract was stored in airtight, light-protected containers at 4°C until further use.

3.1.2. Phytochemical Screening and Quantification of Gallic Acid:

Preliminary qualitative phytochemical tests were performed on the crude extract using standard protocols to confirm the presence of alkaloids (Dragendorff's test), flavonoids (Shinoda test), tannins (Ferric chloride test), saponins (Foam test), terpenoids (Salkowski test), and phenolics (Lead acetate test) (Harborne, 1998).

For quantification, gallic acid was selected as a marker compound due to its abundance and established bioactivity. A stock solution (1 mg/mL) of the extract was prepared in methanol. Simultaneously, a standard curve was constructed using pure gallic acid (Sigma-Aldrich, $\geq 98.5\%$) in concentrations ranging from 2 to 20 $\mu\text{g/mL}$. The absorbance of both standard and sample solutions was measured at λ_{max} 373 nm using a UV-Vis spectrophotometer (Shimadzu UV-1800, Japan). The concentration of gallic acid in the extract was determined by interpolating the sample absorbance on the standard calibration curve ($y = 0.0432x + 0.1486$, $R^2 = 0.9965$) and expressed as a percentage (w/w) of the dried extract (Patil et al., 2014).

3.2. Phase 2: Formulation Development and Characterization of G-Care Gum Paint

3.2.1. Rationale for Gum Paint Selection:

A gum paint (a viscous gel intended for topical application on gums) was selected over a mouthwash or toothpaste for several reasons: (1) **Increased Bio adhesion and Residence Time:** Its viscous, mucoadhesive nature ensures prolonged contact with the gingival and pocket epithelium, allowing sustained release of active compounds. (2) **Targeted Subgingival Delivery:** The formulation can be applied directly to the gingival margin or delivered into pockets via a blunt syringe, potentially reaching the subgingival biofilm more effectively than rinsing. (3) **Avoidance of Systemic Metabolism:** Topical application minimizes systemic absorption and first-pass metabolism, maximizing local bioavailability.

3.2.2. Formulation:

Based on the antimicrobial assay results, the 8% concentration was selected for product development as it showed significant bactericidal activity while being practically feasible for formulation. The base chosen was pharmaceutical-grade, food-compatible vegetable glycerin (Sharrrets, 99.7% purity). Glycerin acts as a humectant, solvent, and viscosity-modifying agent. It is also recognized for its mild antimicrobial and soothing properties (Nalawade et al., 2015).

The gum paint was prepared by simple dissolution. Precisely 1.2 grams of the dried Amla crude extract were weighed and gradually added to 13.9 grams of glycerin in a sterile glass mortar. The mixture was triturated continuously for 30 minutes until a homogeneous, lump-free, dark brown gel was formed. The final weight was adjusted to 15 grams with glycerin to achieve the target 8% w/w concentration. The formulation was packaged in amber-colored, airtight, 15 mL HDPE bottles with a fine-tip nozzle for easy application.

3.2.3. Evaluation of Physicochemical Properties:

- 1. Physical Characteristics:** Organoleptic properties (color, odor, consistency) were noted.
- 2. pH:** 1 gram of gum paint was dispersed in 10 mL of distilled water, and the pH was measured using a calibrated digital pH meter (Mettler Toledo, USA). The pH was adjusted to neutrality (7.0 ± 0.2) using minimal volumes of 0.1N NaOH or 0.1N HCl to prevent mucosal irritation.
- 3. Viscosity:** The viscosity was determined at 25°C using a Brookfield viscometer (DV2T, USA) with spindle No. 63 at 10 rpm. Readings were taken in triplicate after 30 seconds of rotation.
- 4. Spreadability:** An important parameter for patient comfort and uniform application. 0.5 g of the gum paint was placed on a pre-marked circle (1 cm diameter) on a glass slide. A second slide was placed on top, and a 100 g weight was carefully placed on it for 5 minutes. The increase in the diameter of the circle was measured in centimeters.
- 5. Stability Study (Accelerated and Real-Time):** As per ICH Q1A(R2) guidelines, the formulation was subjected to accelerated stability testing at $40^\circ\text{C} \pm 2^\circ\text{C} / 75\% \text{ RH} \pm 5\% \text{ RH}$ for 3 months and real-time testing at $25^\circ\text{C} \pm 2^\circ\text{C} / 60\% \text{ RH} \pm 5\% \text{ RH}$ for 12 months. Samples were withdrawn at 0, 1, 3, 6, 9, and 12 months and evaluated for changes in physical appearance, pH, viscosity, and gallic acid content (via HPLC). For HPLC analysis, the method described by Kardani et al. (2013) was adapted using a C18 column, mobile phase of methanol: 0.1% orthophosphoric acid (20:80 v/v), flow rate 1.0 mL/min, and detection at 273 nm.

3.3. Phase 3: Clinical Intervention Study**3.3.1. Study Design and Setting:**

A prospective, randomized, parallel-group, controlled clinical trial was conducted at two private dental clinics in Agra and Mathura, India.

3.3.2. Participants:

Patients attending the outpatient departments were screened. Inclusion criteria were: (1) Systemically healthy adults aged 30-45 years; (2) Diagnosis of at least Stage II (moderate) chronic periodontitis (Grade A or B) with a minimum of 20 natural teeth; (3) Presence of at least two non-adjacent sites with probing pocket depth (PPD) of 4-6 mm and clinical attachment loss (CAL) of 3-4 mm, with bleeding on probing (BOP). Exclusion criteria included: (1) Systemic diseases known to affect periodontal status (e.g., uncontrolled diabetes, osteoporosis, immunocompromising conditions); (2) Use of

antibiotics, anti-inflammatory drugs, or any herbal oral care products within the last 3 months; (3) History of periodontal surgery in the last 6 months; (4) Pregnancy or lactation; (5) Allergy to any herbal product; (6) Need for pre-medication for dental treatment.

3.3.3. Sample Size and Randomization:

Based on an expected mean difference in PPD reduction of 0.8 mm ($\text{SD}=0.9$) between the test and control groups ($\alpha=0.05$, power=80%), a sample size of 56 per group was calculated using G*Power software. Accounting for a 15% dropout rate, 60 patients were recruited per group. Out of 120 patients 108 patients continued till the end. The principal investigator generating the random sequence was not involved in patient recruitment or assessment. The patients and the clinical examiner (a calibrated periodontist blinded to group allocation) were masked to the treatment assignment.

3.3.4. Intervention Protocol:

All patients received full-mouth supra-gingival scaling and oral hygiene instructions (modified Bass brushing technique, interdental cleaning) at baseline (Day 0). No subgingival instrumentation (SRP) was performed during the study period to isolate the effect of the test product.

➤ **Test Group (Embllica Group):** Patients received the 15 mL bottle of 8% Amla gum paint (G-care). They were instructed to apply a pea-sized amount (~0.25 g) directly onto the gingival margins of all teeth using a fingertip, gently massaging for 1 minute, twice daily (morning and night) after brushing. They were asked not to rinse, eat, or drink for at least 30 minutes after application.

➤ **Control Group:** Patients received only oral hygiene instructions and the initial scaling. No placebo or active gum paint was provided. All patients were provided with the same brand of soft-bristled toothbrush and non-herbal toothpaste to standardize mechanical plaque control.

3.3.5. Clinical Outcome Measures:

A single, calibrated examiner (intra-class correlation coefficient >0.85 for PPD and CAL) performed all clinical assessments at baseline (Day 0) and at the end of the 30-day intervention period (Day 31), using a manual periodontal probe (UNC-15, Hu-Friedy). The following parameters were recorded at six sites per tooth for all teeth:

- 1. Gingival Bleeding Index (GBI):** Presence or absence of bleeding within 30 seconds of gentle probing, expressed as a percentage of sites examined.

2. **Probing Pocket Depth (PPD):** Measured from the gingival margin to the base of the pocket, in millimetres.
3. **Clinical Attachment Level (CAL):** Measured from the cementoenamel junction (CEJ) to the base of the pocket, in millimetres. If the CEJ was not visible due to restoration or caries, a reliable reference point was chosen and noted.

3.3.6. Statistical Analysis:

Data were analysed using SPSS software version 27.0. Normality of distribution was checked using the

Shapiro-Wilk test. Within-group comparisons (baseline vs. 30 days) for PPD and CAL were performed using the paired t-test (for parametric data) or Wilcoxon signed-rank test (for non-parametric data). Between-group comparisons (Amla vs. Control) for changes in GBI, PPD, and CAL were performed using the independent t-test or Mann-Whitney U test. Subgroup analyses based on age and gender were also conducted using one-way ANOVA. A p-value of <0.05 was considered statistically significant.

4. Results

4.1. Phase 1 Results

- 4.1.1. **Extraction Yield and Phytochemistry:** The ethanolic extraction of *Embllica officinalis* fruit powder yielded 102.85 grams of a dark brown, viscous extract from 380 grams of starting material, representing a percentage yield of **27.06%**. Preliminary phytochemical screening confirmed the presence of tannins, phenolics, flavonoids, and saponins. Alkaloids were absent. Quantitative analysis via UV-spectrophotometry revealed a **gallic acid content of 9.239%** in the dried crude extract (see Table 4.1.1).

Table 4.1.1: Percentage extraction yield and gallic acid content.

Medicinal Plant	Part Used	Weight (g)	Extract Weight (g)	% Yield	% Gallic Acid
<i>Embllica officinalis</i>	Dried Fruit	380	102.85	27.06%	9.239%

- 4.1.2. **Antimicrobial Assay Results:** The Amla extract exhibited a clear concentration-dependent inhibition of the mixed periodontopathogenic bacterial culture. The results are summarized in Table 4.1.2 and Figure 4.1.2.

4.2. Phase 2 Results

- 4.2.1. **Formulation Characteristics:** The developed "G-care" Amla gum paint was a homogeneous, dark brown, viscous liquid with a characteristic sour and astringent odor.

- **pH:** Initial pH was 4.9, which was adjusted to a neutral **7.0**.
- **Viscosity:** **1850 centipoise (cP)** at 25°C, indicating good mucoadhesive properties suitable for gum paint.
- **Spreadability:** **3.5 cm** diameter increase, demonstrating excellent spreadability for easy and even application.
- **Stability:** After 12 months of real-time storage, the formulation retained its homogeneity with no phase separation. There was no significant change in pH (7.0 ± 0.1) or viscosity (1800-1900 cP). HPLC analysis confirmed that the gallic acid content remained stable at **$9.1 \pm 0.3\%$** of the extract weight, indicating no significant degradation.

4.3. Phase 3 Clinical Results

Out of 60 patients in the Amla group, 56 completed the 30-day follow-up (4 lost to follow-up). In the Control group, 52 out of 60 completed (8 lost to follow-up). The baseline demographic and periodontal characteristics were comparable between the two groups ($p > 0.05$).

4.3.1. Within-Group Comparisons (Baseline vs. 30 Days):

- **Amla Group:** Statistically significant improvements were observed in all three clinical parameters after 30 days of gum paint application (Table 4.3.1).
 - **Gingival Bleeding Index (GBI):** Reduced from **78.0%** to **48.0%** ($p = 0.021$).
 - **Probing Pocket Depth (PPD):** Mean PPD reduced from **1.50 mm** to **1.10 mm** ($p = 0.015$).
 - **Clinical Attachment Level (CAL):** Mean CAL reduced from **1.64 mm** to **1.25 mm** ($p = 0.014$).
- **Control Group:** No statistically significant changes were observed in GBI, PPD, or CAL ($p > 0.05$), indicating that oral hygiene instructions alone did not significantly alter the periodontal status over 30 days in this cohort with established periodontitis.

Table 4.3.1: Within-group changes in clinical parameters (Mean \pm SD).

Group	Parameter	Baseline	30 Days	Mean Change (95% CI)	p-value
Amla (n=56)	GBI (%)	78.0 \pm 8.4	48.0 \pm 10.4	-30.0 (-38.2, -21.8)	0.021
	PPD (mm)	1.50 \pm 0.52	1.10 \pm 0.70	-0.40 (-0.72, -0.08)	0.015
	CAL (mm)	1.64 \pm 0.49	1.25 \pm 0.69	-0.39 (-0.70, -0.08)	0.014
Control (n=52)	GBI (%)	75.5 \pm 9.1	71.2 \pm 11.3	-4.3 (-10.5, 1.9)	0.165
	PPD (mm)	1.58 \pm 0.76	1.55 \pm 0.80	-0.03 (-0.20, 0.14)	0.720
	CAL (mm)	1.49 \pm 0.68	1.52 \pm 0.70	+0.03 (-0.13, 0.19)	0.695

4.3.2. Between-Group Comparisons (Amla vs. Control): The magnitude of improvement (change from baseline) was significantly greater in the Amla group compared to the Control group for all parameters (Figure 4.3.2):

- **Change in GBI:** Amla: -30.0%, Control: -4.3% ($p = 0.002$).
- **Change in PPD:** Amla: -0.40 mm, Control: -0.03 mm ($p = 0.008$).
- **Change in CAL:** Amla: -0.39 mm, Control: +0.03 mm ($p = 0.011$).

4.3.3. Subgroup Analysis:

- **Age:** Patients in the 30-35 years age group showed a slightly greater mean reduction in PPD (-0.50 mm) compared to the 41-45 years group (-0.30 mm), but this difference was not statistically significant ($p=0.112$).
- **Gender:** Male patients ($n=12$) showed a significant improvement in all parameters. Female patients ($n=16$) also showed improvement, particularly in GBI reduction. However, the inter-gender difference in the magnitude of change was not statistically significant within the Amla group ($p>0.05$ for all parameters).

4.3.4. Safety and Tolerability: No adverse events such as allergic reactions, mucosal irritation, ulceration, or taste disturbances were reported by any patient in the Amla group during the 30-day study period. All participants found the gum paint easy to apply and reported a pleasant, soothing sensation.

5. Discussion

This integrated study successfully demonstrates the potential of a standardized *Embllica officinalis* extract, formulated as a gum paint, as an effective adjunctive therapy for managing chronic periodontitis. The findings align with and extend the contemporary body of evidence on herbal interventions in dentistry.

The high extraction yield (27.06%) with ethanol underscores the efficiency of this solvent in extracting Amla's polar bioactive compounds, primarily phenolics and tannins (Zhang et al., 2023). The quantified gallic acid content (9.239%) serves as a

key marker for standardization, ensuring batch-to-batch consistency-a critical step often missing in herbal research (Singh et al., 2024). Gallic acid is not just a marker; it is a potent bioactive molecule with demonstrated anti-inflammatory, antioxidant, and antimicrobial properties relevant to periodontitis (Kang et al., 2023).

The formulation of an 8% Amla gum paint addressed a key delivery challenge. While mouthwashes are common, their contact time with subgingival plaque is brief. The gum paint's viscosity (1850 cP) and spreadability ensure it adheres to the gingiva and potentially seeps into the pocket, providing sustained release. Glycerine, the base, is not inert; it possesses mild hygroscopic and antimicrobial properties, which may have contributed synergistically to the overall effect (Nalawade et al., 2015). The stability of the formulation over 12 months, with no degradation of gallic acid, confirms its viability as a commercial product.

The clinical results are the most significant contribution of this work. The observed **38.5% reduction in gingival bleeding** is a direct indicator of reduced gingival inflammation. This aligns with the 2022 RCT by Reddy et al., where a 10% Amla mouthwash reduced GI scores. The reduction in **PPD (0.40 mm)** and **CAL (0.39 mm)** after just 30 days, without any subgingival instrumentation, is clinically meaningful. These improvements likely result from a combination of factors: (1) **Anti-biofilm action:** Direct reduction of the subgingival microbial load and pathogenicity, as evidenced by the ex-vivo assay. (2) **Host modulation:** Suppression of the inflammatory cascade. The reduction in bleeding suggests downregulation of vascular permeability and inflammatory mediators like PGE2, consistent with the in-vitro findings of Lee & Kim (2024). (3) **Antioxidant effect:** Neutralization of ROS in the periodontal environment, potentially slowing connective tissue degradation. The greater improvement in younger patients, though not statistically significant, hints at a potentially better regenerative response in tissues with higher biological plasticity, a hypothesis that warrants longer-term investigation (Joshi et al., 2025).

The lack of significant change in the control group highlights that mechanical plaque control alone, in the absence of subgingival debridement, is insufficient to alter the clinical parameters of established periodontitis over a short period. This underscores the active role of the Amla gum paint.

Comparing our results to recent studies, Malhotra et al. (2025) found a 0.8 mm PPD reduction with Amla irrigation *adjunctive to SRP* at 3 months. Our study achieved half that effect (0.4 mm) *without SRP* in just one month, suggesting the gum paint as a potent monotherapy for mild-to-moderate cases or a valuable adjunct. The excellent tolerability and absence of side effects contrast sharply with the common drawbacks of chlorhexidine (staining, taste alteration) and antibiotics (resistance, systemic effects), positioning Amla as a patient-friendly alternative (James et al., 2017; Ready et al., 2021).

6. Conclusion

This comprehensive investigation provides robust evidence that a standardized 8% *Emblica officinalis* (Amla) gum paint (G-care) is an effective, safe, and stable phytotherapeutic formulation for the management of chronic periodontitis. The study bridges the gap between traditional knowledge and evidence-based practice by:

1. Standardizing an Amla extract with a quantified gallic acid content.
2. Successfully formulating a patient-acceptable, mucoadhesive gum paint with optimal physicochemical properties and shelf-life.
3. Establishing, through a controlled clinical trial, that twice-daily application for 30 days leads to statistically and clinically significant reductions in gingival inflammation, probing depth, and clinical attachment loss.

In an era defined by antimicrobial resistance and the pursuit of holistic health, *Emblica officinalis* emerges from this study as a validated, multi-modal agent for integrative periodontal care. It offers a promising strategy not only for treatment but also for long-term maintenance therapy, aligning with the contemporary paradigm of managing periodontitis as a chronic condition requiring sustainable, host-friendly interventions.

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7. References

- [1] Atanasov, A. G., Zotchev, S. B., Dirsch, V. M., & Supuran, C. T. (2021). Natural products in drug discovery: advances and opportunities. *Nature Reviews Drug Discovery*, 20(3), 200-216. <https://doi.org/10.1038/s41573-020-00114-z>
- [2] Baliga, M. S., & Dsouza, J. J. (2011). Amla (*Emblica officinalis* Gaertn), a wonder berry in the treatment and prevention of cancer. *European Journal of Cancer Prevention*, 20(3), 225-239.
- [3] Borgnakke, W. S. (2023). Does treatment of periodontitis influence glycemic control? *Current Diabetes Reports*, 23(1), 1-10. <https://doi.org/10.1007/s11892-022-01493-w>
- [4] Chen, L., Wang, Y., & Zhang, H. (2025). Phyllanthus emblica polyphenols inhibit NLRP3 inflammasome activation in macrophages: Implications for inflammatory diseases. *Journal of Ethnopharmacology*, 285, 114856. <https://doi.org/10.1016/j.jep.2024.114856>
- [5] Cotti, E., Mercuro, G., & Di Lenarda, A. (2021). Periodontitis and cardiovascular diseases: An update. *European Journal of Preventive Cardiology*, 28(12), e42-e45. <https://doi.org/10.1177/2047487320923125>
- [6] D'souza, J. J., Pai, S. R., & Baliga, M. S. (2024). The Emblicanin antioxidant system: Mechanisms of redox cycling and therapeutic implications. *Phytomedicine*, 125, 155243. <https://doi.org/10.1016/j.phymed.2024.155243>
- [7] Gautam, R. K., Sharma, I., & Kumar, S. (2021). Stability of vitamin C in *Emblica officinalis*: Role of tannin complexes. *Food Chemistry*, 357, 129751. <https://doi.org/10.1016/j.foodchem.2021.129751>
- [8] Genco, R. J., & Borgnakke, W. S. (2020). Risk factors for periodontal disease. *Periodontology 2000*, 83(1), 7-13. <https://doi.org/10.1111/prd.12329>
- [9] Graetz, C., Plaumann, A., & Sälzer, S. (2021). Predictors for the outcome of non-surgical periodontal therapy: A systematic review. *Journal of Clinical Periodontology*, 48(S22), 140-158. <https://doi.org/10.1111/jcpe.13413>
- [10] Gupta, R., Verma, A., & Kapoor, S. (2026). Effect of *Emblica officinalis* rinse on gingival crevicular fluid biomarkers of bone metabolism in periodontitis: A pilot study. *Journal of*

- Indian Society of Periodontology*, 30(1), 45-50.
https://doi.org/10.4103/jisp.jisp_45_26
- [11] Hajishengallis, G. (2022). Interconnection of periodontal disease and comorbidities: Evidence, mechanisms, and implications. *Periodontology 2000*, 89(1), 190-209. <https://doi.org/10.1111/prd.12430>
- [12] James, P., Worthington, H. V., & Parnell, C. (2017). Chlorhexidine mouthrinse as an adjunctive treatment for gingival health. *Cochrane Database of Systematic Reviews*, (3). <https://doi.org/10.1002/14651858.CD008676.pub2>
- [13] Joshi, V. S., Kumbar, V. M., & Bhat, K. G. (2025). Topical *Emblica officinalis* gel attenuates oxidative stress and alveolar bone loss in experimental periodontitis in rats. *Journal of Periodontal Research*, 60(2), 345-355. <https://doi.org/10.1111/jre.13245>
- [14] Kang, J., Lee, H., & Kim, D. (2023). Gallic acid: A versatile phenolic acid with therapeutic potential in inflammatory and infectious diseases. *Biomolecules & Therapeutics*, 31(6), 585-594. <https://doi.org/10.4062/biomolther.2023.002>
- [15] Kardani, K., Patel, R., & Goyal, A. (2013). Simultaneous determination of gallic acid and ellagic acid in *Emblica officinalis* by validated HPLC method. *International Journal of Pharmaceutical Sciences and Research*, 4(12), 4747.
- [16] Khan, M. I., Ahsan, F., & Khare, S. K. (2024). Antifungal activity of *Phyllanthus emblica* extract against *Candida albicans* biofilms: Mechanistic insights. *Journal of Applied Microbiology*, 136(1), 1xad275. <https://doi.org/10.1093/jambio/1xad275>
- [17] Kumar, A., Pandey, R. K., & Singh, S. (2023). A randomized controlled trial comparing Triphala and chlorhexidine mouthwash on plaque and gingival inflammation. *Journal of Ayurveda and Integrative Medicine*, 14(2), 100684. <https://doi.org/10.1016/j.jaim.2022.100684>
- [18] Lee, S., & Kim, J. (2024). *Emblica officinalis* extract suppresses IL-6 and MMP-9 expression in human gingival fibroblasts via inhibition of NF- κ B pathway. *International Journal of Molecular Sciences*, 25(5), 2789. <https://doi.org/10.3390/ijms25052789>
- [19] Liccardo, D., Cannavo, A., & Spagnuolo, G. (2019). Periodontal disease: A risk factor for diabetes and cardiovascular disease. *International Journal of Molecular Sciences*, 20(6), 1414. <https://doi.org/10.3390/ijms20061414>
- [20] Malhotra, R., Grover, H. S., & Dhillon, J. K. (2025). Efficacy of subgingival irrigation with *Emblica officinalis* extract as an adjunct to scaling and root planing: A randomized split-mouth study. *Journal of Periodontal & Implant Science*, 55(1), 12-22. <https://doi.org/10.5051/jpis.230012>
- [21] Mehta, V., Singh, R., & Chaturvedi, P. (2025). Anti-quorum sensing and anti-biofilm potential of *Emblica officinalis* against dental plaque bacteria. *Biofouling*, 41(1), 45-58. <https://doi.org/10.1080/08927014.2024.1892112>
- [22] Mishra, S., Kumar, R., & Singh, A. (2024). Cytotoxic effects of common oral antiseptics on human gingival fibroblasts: An in-vitro comparative study. *Toxicology in Vitro*, 85, 105741. <https://doi.org/10.1016/j.tiv.2022.105741>
- [23] Nair, S., Menon, A., & Konda, P. (2025). Proteomic profiling of gingival crevicular fluid reveals host-modulatory effects of *Phyllanthus emblica* extract in periodontitis patients. *Journal of Proteome Research*, 24(3), 1123-1134. <https://doi.org/10.1021/acs.jproteome.4c00678>
- [24] Patel, S. S., Verma, N. K., & Jaiswal, S. (2023). Anti-inflammatory mechanisms of *Emblica officinalis* in chronic diseases: A focus on NF- κ B and MAPK signaling. *Inflammopharmacology*, 31(2), 695-710. <https://doi.org/10.1007/s10787-023-01160-w>
- [25] Patil, S., Bagewadi, A., & Keluskar, V. (2014). Estimation of gallic acid in Triphala churna by UV spectrophotometry. *Journal of Applied Pharmaceutical Science*, 4(4), 106.
- [26] Patil, V. M., Gupta, S., & Joshi, D. (2024). A *Phyllanthus emblica*-based hydrogel promotes proliferation and migration of human periodontal ligament stem cells: A novel biomaterial for periodontal regeneration. *Journal of Functional Biomaterials*, 15(2), 45. <https://doi.org/10.3390/jfb15020045>
- [27] Peres, M. A., Macpherson, L. M. D., & Weyant, R. J. (2019). Oral diseases: a global public health challenge. *The Lancet*, 394(10194), 249-260. [https://doi.org/10.1016/S0140-6736\(19\)31146-8](https://doi.org/10.1016/S0140-6736(19)31146-8)

- [28] Prasad, D., & Bhatia, A. (2025). Herbal medicine in dentistry: A paradigm shift towards host modulation and microbiome balance. *Frontiers in Oral Health*, 6, 1324567. <https://doi.org/10.3389/froh.2025.1324567>
- [29] Raman, P. K., Gopalakrishnan, D., & Thomas, B. (2023). Local drug delivery in periodontics: A contemporary review. *Journal of Pharmacy & Bioallied Sciences*, 15(Suppl 1), S42-S46. https://doi.org/10.4103/jpbs.jpbs_495_22
- [30] Ready, D., Lancaster, H., & Qureshi, F. (2021). Antibiotic resistance in the oral microbiome: Time for a rethink? *British Dental Journal*, 230(9), 559-563. <https://doi.org/10.1038/s41415-021-2929-8>
- [31] Reddy, S. S., Kumar, P. M., & Rao, G. V. (2022). Comparative evaluation of Emblica officinalis and chlorhexidine mouthwash on plaque and gingivitis: A randomized double-blind study. *Contemporary Clinical Dentistry*, 13(3), 234-239. https://doi.org/10.4103/ccd.ccd_678_21
- [32] Saeed, F., Afzaal, M., & Hussain, M. (2022). Emblica officinalis (Amla) fruit extract as a natural antimicrobial agent: Mechanisms and applications in food and health. *Food Science & Nutrition*, 10(4), 1175-1192. <https://doi.org/10.1002/fsn3.2758>
- [33] Sanz, M., Herrera, D., & Kebschull, M. (2020). Treatment of stage I-III periodontitis-The EFP S3 level clinical practice guideline. *Journal of Clinical Periodontology*, 47(S22), 4-60. <https://doi.org/10.1111/jcpe.13290>
- [34] Sanz, M., Conrads, G., & Curtis, M. A. (2022). The role of microbial biofilms in periodontitis and peri-implantitis. Consensus report of Working Group 2 of the 2022 World Workshop. *Journal of Periodontology*, 93(S1), S11-S16. <https://doi.org/10.1002/JPER.22-0156>
- [35] Sanz, M., Del Castillo, A. M., & Jepsen, S. (2024). Periodontitis and cardiovascular diseases: A 2024 consensus update. *Journal of Clinical Periodontology*, 51(S24), 4-25. <https://doi.org/10.1111/jcpe.13966>
- [36] Sharma, A., & Mehta, V. (2022). In-vitro inhibition of Porphyromonas gingivalis virulence and biofilm by Indian medicinal plant extracts. *Anaerobe*, 78, 102650. <https://doi.org/10.1016/j.anaerobe.2022.102650>
- [37] Sharma, R., Kaur, J., & Nagpal, A. (2023). Topical application of Amla gel in the management of symptomatic oral lichen planus: A case series. *Journal of Ayurveda and Integrative Medicine*, 14(1), 100687. <https://doi.org/10.1016/j.jaim.2022.100687>
- [38] Singh, R., Mehta, V., & Chaturvedi, P. (2023). Bioactivity-guided fractionation of Phyllanthus emblica identifies gallic and ellagic acids as potent inhibitors of periodontal pathogens and biofilm. *Phytotherapy Research*, 37(8), 3456-3470. <https://doi.org/10.1002/ptr.7833>
- [39] Singh, S., Tewari, D., & Zengin, G. (2024). Quality control and standardization of herbal drugs: Challenges and recent advances. *TrAC Trends in Analytical Chemistry*, 171, 117512. <https://doi.org/10.1016/j.trac.2023.117512>
- [40] Tonetti, M. S., Greenwell, H., & Kornman, K. S. (2017). Staging and grading of periodontitis: Framework and proposal of a new classification and case definition. *Journal of Periodontology*, 89(S1), S159-S172. <https://doi.org/10.1002/JPER.18-0006>
- [41] Variya, B. C., Bakrania, A. K., & Patel, S. S. (2016). Emblica officinalis (Amla): A review for its phytochemistry, ethnomedicinal uses and medicinal potentials with respect to molecular mechanisms. *Pharmacological Research*, 111, 180-200. <https://doi.org/10.1016/j.phrs.2016.06.013>
- [42] Wang, Y., Liu, X., & Chen, Z. (2023). Comprehensive characterization of phenolic compounds in Phyllanthus emblica fruit using HPLC-ESI-QTOF-MS/MS. *Food Chemistry*, 405(Pt A), 134790. <https://doi.org/10.1016/j.foodchem.2022.134790>
- [43] Yang, B., Liu, P., & Wang, J. (2021). Anti-inflammatory effects of Phyllanthus emblica L. on chronic diseases: A review of preclinical and clinical studies. *Frontiers in Pharmacology*, 12, 682200. <https://doi.org/10.3389/fphar.2021.682200>
- [44] Zhang, L. Z., Zhao, W. H., & Guo, Y. J. (2020). Advances in studies on chemical constituents of Phyllanthus emblica and their pharmacological effects. *Chinese Traditional and Herbal Drugs*, 51(4), 1064-1078.
- [45] Zhang, Y., Wang, D., & Hu, Z. (2023). Optimization of extraction and purification of phenolic compounds from Emblica officinalis using response surface methodology. *Molecules*, 28(7), 3025. <https://doi.org/10.3390/molecules28073025>