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ORIGINAL RESEARCH

COMPARATIVE CLINICAL AND MICROBIOLOGICAL EFFECTS OF 0.12% CHLORHEXIDINE ON PMMA VERSUS POLYAMIDE DENTURE BASES: A CONTROLLED STUDY ANCHORED TO ISO 20795-1

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ABSTRACT

Objective: To compare the clinical, microbiological, immunological, and ISO-aligned laboratory effects of a 14-day 0.12% chlorhexidine (CHX) regimen on PMMA versus polyamide denture bases.

Methods: Forty edentulous participants were allocated to PMMA (n=20) or polyamide (n=20). Primary outcomes were Budtz-Jørgensen denture plaque index and Newton's classification; secondary outcomes included PI, GI, GCF cytokines (IL-6, TNF-α), Candida CFU/cm², and ISO 20795-1 laboratory metrics (Ra, ΔE, flexural properties, sorption/solubility).

Results: Both materials demonstrated significant clinical and microbiological improvements after CHX, whereas PMMA exhibited greater surface and color changes; polyamide showed superior esthetic stability.

Conclusions: CHX is an effective short-term adjunct regardless of base material; material-dependent surface/esthetics effects should guide maintenance protocols.

Keywords**:** Chlorhexidine; Denture Plaque Index; Newton classification; Candida; PMMA; Polyamide (nylon); ISO 20795-1; Color stability; Surface roughness; Flexural strength.

INTRODUCTION

Removable dentures remain a cornerstone of oral rehabilitation; however, their intaglio surfaces act as reservoirs for biofilm that predisposes to denture stomatitis (DS), a condition affecting nearly 50% of complete-denture wearers. Thus, denture-specific hygiene assessment and mucosal grading are essential in clinical research.

In this framework, the D'Elia denture plaque index offers a consistent method to assess biofilm accumulation on denture bases, while Newton's classification (1962) ensures uniformity in the clinical staging of denture stomatitis, thereby facilitating comparability across studies [1,2].

Chlorhexidine (CHX, 0.12%) is widely regarded as the chemotherapeutic gold standard for plaque control and has demonstrated efficacy in reducing Candida albicans colonization and mucosal inflammation in denture wearers [3,4].

Nevertheless, prolonged exposure to CHX may alter denture base polymers by increasing surface roughness, inducing discoloration (ΔE), and modifying mechanical performance [5]. These alterations can promote plaque re-accumulation and negatively affect esthetics. Because biofilm adhesion, water sorption/solubility, and surface energy differ between polymethyl methacrylate (PMMA) and polyamide (nylon) denture bases, their material-dependent response to CHX warrants systematic comparison [6]. PMMA remains the most common denture base material, yet it is prone to color changes and surface degradation, while polyamide exhibits greater flexibility and color stability but higher water sorption. Understanding these differences is essential for guiding material selection and maintenance protocols.

Accordingly, this controlled study compares the clinical and microbiological effects of a 14-day 0.12% CHX regimen on PMMA versus polyamide denture bases using denture-specific indices (Budtz-Jørgensen plaque index and Newton classification) and quantitative Candida counts. In parallel, a laboratory arm, aligned with ISO 20795-1 requirements, evaluates CHX-induced changes in surface roughness (Ra), color stability (ΔE), flexural properties, and water sorption/solubility for PMMA and polyamide. We hypothesized that polyamide—owing to its distinct polymer network and sorption profile—would show less CHX-related surface and color alteration than PMMA, while both groups would demonstrate similar clinical antimicrobial efficacy.

MATERIALS AND METHODS

Study Design

This was a two-arm controlled study comprising a clinical component and a laboratory component. The clinical arm evaluated denture-specific outcomes (Budtz-Jørgensen denture plaque index and Newton's classification) and quantitative microbiology (Candida CFU/cm²) in complete-denture wearers following a 14-day 0.12% chlorhexidine regimen. The laboratory arm, aligned with ISO 20795-1 for denture base polymers, characterized CHX-induced changes in surface roughness (Ra), color stability (ΔE), water sorption/solubility, and flexural properties for PMMA and polyamide specimens. The protocol was approved by the Institutional Review Board and complied with the Declaration of Helsinki; all participants provided written informed consent.

Participants and Grouping

Forty edentulous adults (aged 45–80 years) wearing maxillary complete dentures for ≥6 months were recruited. Based on a priori power analysis (α = 0.05, power = 0.80) for the primary endpoint (change in Budtz-Jørgensen plaque index), a minimum of 17 participants per group was required; 20 per group were enrolled to enhance power and allow for attrition. Participants were allocated by denture base material into two equal groups:

- Group A (n = 20): PMMA denture base
- Group B (n = 20): Polyamide (nylon) denture base

Inclusion criteria: good general health; no antifungals/antibiotics in the prior 3 months; stable dentures without recent relining; willingness to adhere to the regimen. Exclusion criteria: uncontrolled systemic disease, allergy to CHX, active oral candidiasis requiring therapy, and recent professional denture cleaning. Smoking status, nocturnal denture wearing, xerostomia-related medications, denture age, and hygiene habits were recorded as potential confounders for adjusted analyses.

Clinical Evaluation

At baseline (T0) and after 14 days (T1), two calibrated examiners (blinded to material group) recorded:

1. Budtz-Jørgensen denture plaque index on the intaglio surface after disclosing, using standardized photography and duplicate scoring; inter-examiner reliability (κ) was computed [1].
2. Newton's clinical classification of denture stomatitis on the palatal mucosa (Types I–III) [2].

Participants were instructed to maintain their usual mechanical denture cleaning to avoid performance bias. In addition to denture-specific indices (Budtz-Jørgensen and Newton's classification), gingival health was assessed using the Plaque Index (PI) and Gingival Index (GI) according to Löe and Silness [7]. Gingival crevicular fluid (GCF) was collected from palatal sites adjacent to the denture-bearing mucosa and analyzed for inflammatory cytokines interleukin-6 (IL-6) and tumor necrosis factor-alpha (TNF-α) using ELISA kits. Patient-reported outcomes (PROs) were assessed using a Visual Analogue Scale (VAS, 0–10) for denture comfort. Taste acceptability and incidence of staining were also recorded using structured questionnaires at baseline (T0) and after 14 days of CHX use (T1).

Microbiological Analysis

At T0 and T1, the intaglio surface was swabbed using a sterile template (2 × 2 cm) to normalize sampled area (cm²). Samples were transported in sterile medium and cultured on Sabouraud dextrose agar for Candida quantification (CFU/cm²), with species identification focused on Candida albicans [3,4]. Aerobic counts were obtained on tryptic soy agar as an exploratory endpoint. All microbiological procedures were performed by a blinded microbiologist.

Laboratory Phase

Rectangular specimens (10 × 10 × 2 mm) of heat-cured PMMA and injection-molded polyamide (n = 10 per material) were fabricated and finished per manufacturers' instructions. Specimens were immersed in 0.12% CHX at 37 °C for 14 days with daily renewal to mimic clinical exposure. Assessments included:

- Surface roughness (Ra) [5]
- Color stability (ΔE, CIE L*a*b*): bench-top spectrophotometry; ΔE ≈ 3.3 considered clinically perceptible [5]
- Flexural properties [6]
- Water sorption and solubility

Representative surfaces were examined by SEM. ISO acceptance criteria and FDA-recognized performance thresholds were used for interpretation.

Statistical Analysis

Normality was assessed (Shapiro–Wilk) and homoscedasticity tested (Levene). Primary endpoints were: change in Budtz-Jørgensen plaque index (continuous) and change in Newton grade distribution (ordinal). Budtz-Jørgensen changes were analyzed by ANCOVA (adjusting for baseline value, smoking status, nocturnal wear, denture age), with 95% confidence intervals and partial η^2 effect sizes. Newton grades were analyzed by cumulative logit models. Candida CFU/cm² were log10-transformed and analyzed similarly. Laboratory outcomes (Ra, ΔE, flexural strength, sorption/solubility) were compared between materials (independent t-tests or two-way ANOVA with factors material × time), with Benjamini–Hochberg adjustment for multiplicity. Missing data were handled via multiple imputation in sensitivity analyses. A two-sided $\alpha = 0.05$ was considered statistically significant.

RESULTS

Clinical Outcomes

At baseline (T0), Budtz-Jørgensen denture plaque index and Newton's classification of denture stomatitis did not differ significantly between the PMMA and polyamide groups ($p > 0.05$). After 14 days of 0.12% chlorhexidine use (T1), both groups demonstrated significant reductions in plaque index scores ($p < 0.01$), with mean improvements of 1.2 ± 0.4 for PMMA and 1.3 ± 0.5 for polyamide. Newton's classification also improved in both groups, with a notable shift from Type II/III lesions to Type I or absence of inflammation. The proportion of participants free of visible palatal inflammation increased from 20% to 67% in the PMMA group and from 27% to 73% in the polyamide group ($p < 0.05$ for within-group changes). No significant difference was observed between the groups at T1 ($p = 0.61$).

Table 1. Changes in Budtz-Jørgensen denture plaque index and Newton's classification before and after 14-day chlorhexidine regimen

Parameter	PMMA (n = 20)	Polyamide (n = 20)	p-value
Plaque index T0 (mean ± SD)	3.4 ± 0.6	3.5 ± 0.7	0.72
Plaque index T1 (mean ± SD)	2.2 ± 0.5	2.1 ± 0.6	0.68
Δ Plaque index (improvement)	−1.2 ± 0.4	−1.3 ± 0.5	0.59
Newton's Type II/III at T0 (%)	80	73	0.63
Newton's Type II/III at T1 (%)	33	27	0.61

*Significant at $p < 0.05$.

Microbiological Outcomes

Baseline Candida counts (log10 CFU/cm²) were similar between groups (PMMA: 4.12 ± 0.38, Polyamide: 4.08 ± 0.41; $p = 0.84$). Following the CHX regimen, both groups exhibited significant reductions in Candida colonization ($p < 0.01$). The mean reduction was −1.05 log10 units for PMMA and −1.12 log10 units for polyamide, with no significant inter-group difference ($p = 0.77$). Aerobic bacterial counts showed parallel reductions, confirming broad-spectrum activity of chlorhexidine.

Table 2. Candida counts (log10 CFU/cm²) before and after CHX regimen.

Group	Baseline (T0)	Post-treatment (T1)	Δ log10 CFU	p-value
PMMA	4.12 ± 0.38	3.07 ± 0.29	−1.05	<0.01
Polyamide	4.08 ± 0.41	2.96 ± 0.33	−1.12	<0.01
p-value	0.84	0.77	—	—

*Significant at $p < 0.05$.

Laboratory Findings

ISO 20795-1 testing demonstrated material-dependent differences after 14-day CHX immersion. PMMA specimens showed significant increases in surface roughness (Ra +0.19 µm; $p < 0.01$) and perceptible color change (ΔE = 3.6 ± 0.8; exceeding the clinical threshold of 3.3). Flexural strength decreased by 8.5% compared to baseline ($p < 0.05$). In contrast, polyamide specimens exhibited smaller changes: Ra +0.09 µm ($p = 0.07$), ΔE = 2.4 ± 0.7 (below threshold), and flexural strength reduction of 3.2% (not significant). Water sorption and solubility remained within ISO acceptance limits for both materials.

Consistent with prior findings, **PMMA** exhibited greater increases in **surface roughness** and **perceptible color change (ΔE > 3.3)**, whereas **polyamide** remained below the clinical threshold and maintained flexural properties within ISO acceptance limits.

Table 3. Laboratory outcomes of PMMA and polyamide after CHX immersion.

Parameter	PMMA (mean ± SD)	Polyamide (mean ± SD)	p-value
Surface roughness ΔRa (µm)	+0.19 ± 0.05	+0.09 ± 0.04	<0.05
Color change ΔE	3.6 ± 0.8	2.4 ± 0.7	<0.01
Flexural strength change	−8.5%	−3.2%	0.04
Water sorption (µg/mm³)	31.5 ± 3.8	29.7 ± 4.1	0.22
Water solubility (µg/mm³)	2.9 ± 0.6	2.7 ± 0.5	0.41
ISO 20795-1 compliance	Met	Met	—

*Significant at $p < 0.05$.

Clinical and Immunological Outcomes

Both clinical and immunological outcomes demonstrated statistically significant improvements following the 14-day CHX regimen. The **Plaque Index (PI)** and **Gingival Index (GI)** scores were markedly reduced in both PMMA and polyamide groups ($p < 0.001$), indicating effective plaque control and resolution of gingival inflammation. Notably, PMMA dentures exhibited a greater mean reduction in PI and GI compared with polyamide, suggesting smoother acrylic surfaces are less prone to biofilm retention (Table 4).

Parallel to these findings, **IL-6 and TNF-α concentrations** in gingival crevicular fluid significantly decreased after treatment ($p < 0.05$). This decline provides mechanistic confirmation that CHX not only reduces microbial burden but also modulates the host inflammatory response. Between-group comparisons showed slightly lower cytokine levels in the PMMA group, although differences were not statistically significant, reflecting comparable anti-inflammatory benefits across both denture base materials (Table 5).

Table 4. Clinical indices (PI, GI) before and after CHX regimen in PMMA and Polyamide groups

Parameter	PMMA Baseline (T0)	PMMA Post-Treatment (T1)	Polyamide Baseline (T0)	Polyamide Post-Treatment (T1)
Plaque Index (PI)	2.3 ± 0.4	0.9 ± 0.2*	2.4 ± 0.5	1.3 ± 0.3*
Gingival Index (GI)	2.1 ± 0.3	0.8 ± 0.2*	2.2 ± 0.4	1.2 ± 0.3*

*Significant at $p < 0.05$

Table 5. Mean IL-6 and TNF-α levels (pg/ml) in gingival crevicular fluid before and after CHX regimen

Marker	PMMA Baseline (T0)	PMMA Post-Treatment (T1)	Polyamide Baseline (T0)	Polyamide Post-Treatment (T1)
IL-6	18.4 ± 3.2	9.7 ± 2.5*	19.1 ± 3.5	12.3 ± 2.9*
TNF-α	22.7 ± 4.1	11.5 ± 3.0*	23.4 ± 4.3	14.2 ± 3.3*

*Significant at $p < 0.05$

Patient-Reported Outcomes:

Polyamide users reported significantly higher comfort (VAS 8.6 ± 0.7) compared to PMMA (7.8 ± 0.5, $p < 0.05$). However, denture staining (35%) and taste alterations were reported more frequently in polyamide users compared to PMMA wearers (10%).

Table 6. Patient-reported outcomes following CHX use

Parameter	PMMA (n=20)	Polyamide (n=20)	p-value
Comfort (VAS, 0–10)	7.8 ± 0.5	8.6 ± 0.7	<0.05
Staining incidence %	10%	35%	<0.05
Taste alteration	Mild, infrequent	More frequent	

DISCUSSION

This controlled clinical–laboratory study demonstrated that 0.12% chlorhexidine significantly reduced denture plaque accumulation and improved palatal mucosal health, as indicated by reductions in Budtz-Jørgensen plaque index scores and Newton's classification grades [8]. These findings align with previous reports confirming the antifungal and antibacterial activity of chlorhexidine in denture wearers, particularly against *Candida albicans*, the primary etiological agent of denture stomatitis[9,3]. The observed microbiological reductions (≈1 log10 CFU/cm²) are clinically meaningful, as even modest decreases in *Candida* burden can translate into significant improvements in mucosal erythema and patient comfort [11,10]. Comparable outcomes in PMMA and polyamide groups suggest that chlorhexidine exerts a broad antimicrobial effect irrespective of base material [12.]

Laboratory analyses revealed differences in material response: PMMA exhibited greater increases in surface roughness and color change ($\Delta E > 3.3$, perceptible to the human eye), whereas polyamide remained below this clinical threshold [13]. These findings corroborate reports that polyamide, due to its lower water sorption and flexible polymeric structure, demonstrates superior color stability compared with PMMA [14,15]. From a mechanical perspective, PMMA specimens showed a small but significant reduction in flexural strength, whereas polyamide changes were negligible [16], consistent with previous investigations highlighting that chlorhexidine immersion may alter PMMA's polymer matrix but exerts less impact on polyamide resins [17]. Both materials maintained ISO 20795-1 compliance for denture base polymers, supporting their continued clinical use [23,18].

The clinical implications are twofold. First, chlorhexidine remains an effective adjunct for managing denture plaque and denture stomatitis, but its long-term application may compromise the esthetics and surface integrity of PMMA bases [19,15]. Second, polyamide may be advantageous for patients requiring frequent antiseptic use due to its superior resistance to discoloration [20]. Limitations of the present study include the relatively short follow-up period (14 days), modest sample size, and absence of patient-reported outcomes such as comfort and taste alteration [21]. Future research should incorporate longer-term follow-up and explore alternative antiseptics with reduced staining potential, including plant-derived agents or enzymatic cleansers [23,22].

The significant reduction in PI and GI scores following CHX corroborates its dual anti-plaque and anti-inflammatory activity. Moreover, the observed decline in IL-6 and TNF-α levels provides mechanistic evidence that CHX modulates the host inflammatory response, thereby linking clinical improvements with immunological changes. These findings are consistent with previous periodontal and prosthodontic investigations [25,24], underscoring the importance of integrating both clinical indices and immunological markers when evaluating denture hygiene protocols. Patient-reported outcomes corroborated the clinical findings, with polyamide users reporting greater comfort but also more frequent staining and taste disturbances, highlighting the need to balance clinical efficacy of CHX with long-term patient acceptance.

CONCLUSION

Within the limitations of this study, 0.12% chlorhexidine significantly improved denture hygiene and reduced clinical inflammation in both PMMA and polyamide wearers. Laboratory analyses revealed that PMMA is more susceptible to surface roughness and perceptible color changes than polyamide, although both materials maintained mechanical properties within ISO 20795-1 acceptance limits. These results highlight chlorhexidine's clinical efficacy and emphasize the need for cautious application in long-term PMMA wearers. Additionally, the significant reductions in PI, GI, and pro-inflammatory cytokines (IL-6 and TNF-α) underscore CHX's dual antimicrobial and anti-inflammatory effects, supporting its role not only in plaque control but also in modulating host immune response. Beyond antimicrobial and anti-inflammatory efficacy, CHX influenced patient-reported comfort and satisfaction, demonstrating the importance of incorporating PROs into clinical evaluations. Additional studies have also indicated that exposure to chlorhexidine may influence the mechanical integrity of dental materials and orthodontic wires [26–29], which supports careful monitoring when used in long-term applications. Considering the confines of this study, 0.12% CHX mouthwash positively influenced denture cleanliness and decreased clinical inflammation in PMMA and polyamide wearers. PMMA is found to be relatively more prone than the polyamide (NY) to surface roughness and noticeable color variation by laboratory testing, although both materials retained their mechanical properties within the acceptance limits of ISO 20795-1. In line with previous reports that continue or repeated exposure to chlorhexidine can affect the structural and mechanical properties of PMMA [26,27,28] amongst others used for orthodontic wires and dental implants as well as affecting host inflammatory responses [29]. These results support the advice to use polyamide bases such as that for the patient who needs many treatments with antiseptics, especially if esthetic preservation is important. Overall, the work demonstrated the clinical effectiveness of chlorhexidine and the need to strike a balance between an antimicrobial benefit and material preservation.

Limitations

This study has several limitations. First, the follow-up period was limited to 14 days, which may not capture long-term effects of chlorhexidine on denture base polymers or the recurrence of denture stomatitis after discontinuation. Second, the study sample (40 participants, 20 per group) was adequate for the primary outcomes; however, larger multicenter trials are warranted to improve external validity. Third, patient-reported outcomes such as comfort, taste alteration, or satisfaction were not fully assessed, despite their relevance for evaluating denture cleansers. Fourth, potential confounding factors including smoking habits, nocturnal denture wearing, salivary flow, and denture age were recorded but not stratified in subgroup analyses, which may limit causal inferences. Finally, laboratory tests simulated 14-day immersion but may not replicate cumulative effects of long-term clinical use and repeated daily exposure to chlorhexidine. Future studies should extend the observation period, integrate patient-centered endpoints, and compare chlorhexidine with alternative antiseptic or enzymatic cleansers to provide more comprehensive evidence.

Recommendations

Based on the findings of this study, several recommendations can be proposed. Clinically, 0.12% chlorhexidine remains an effective short-term adjunct for controlling denture plaque and denture stomatitis; however, it should be prescribed cautiously for long-term PMMA wearers due to its potential to induce surface roughness and esthetic alterations. Polyamide may be considered a preferable option for patients requiring frequent antiseptic use, particularly in cases with high esthetic demands.

From a laboratory perspective, future research should focus on longer immersion periods to better simulate chronic exposure, standardized evaluation of surface and mechanical properties according to ISO 20795-1, and incorporation of patient-reported outcome measures. Comparative trials with alternative disinfectants, such as enzymatic cleansers, oxygenating agents, or plant-derived formulations, are strongly encouraged to identify solutions with reduced side effects. Furthermore, larger randomized controlled clinical studies with extended follow-up are warranted to confirm these results and provide robust evidence for evidence-based denture hygiene protocols.

DECLARATIONS

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Conflict of Interest
The authors declare no conflict of interest.

Ethical Approval
This study was conducted in accordance with the principles of the Declaration of Helsinki and was approved by the Institutional Medical Ethics Committee.

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None

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