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Original Research Article

Comparative evaluation of the microbial load and dimensional accuracy of dental impressions disinfected with immersion under 2% glutaraldehyde and 0.5% sodium hypochlorite: An ex vivo study

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Abstract

Background: Dental impressions are potential vectors for cross-infection due to contamination with saliva and blood. Effective disinfection is critical to prevent transmission of pathogens without compromising the dimensional accuracy of the impressions. This study aimed to evaluate and compare the antimicrobial efficacy of 2% glutaraldehyde and 0.5% sodium hypochlorite on irreversible hydrocolloid (alginate) and addition silicone impression materials and to assess their impact on dimensional stability.

Materials and Methods: An ex vivo study was conducted using impressions obtained from systemically healthy patients. A total of 180 impressions (90 alginate and 90 addition silicone) were randomly assigned to three subgroups (15 samples each): Group A – no disinfection (control), Group B – immersion in 2% glutaraldehyde, and Group C – immersion in 0.5% sodium hypochlorite, each for 10 minutes. Dimensional stability was assessed by measuring inter-point distances on resultant stone casts using digital vernier calipers. Microbial analysis was performed by culturing disinfected impressions on nutrient, blood, and MacConkey agars to evaluate colony-forming units (CFU/cm²) and identify microbial strains.

Results: No statistically significant differences in dimensional measurements were observed across disinfected and control groups ($p > 0.05$), indicating preservation of dimensional stability. Both disinfectants significantly reduced microbial counts compared to controls ($p < 0.001$), with sodium hypochlorite showing slightly greater antimicrobial efficacy. *Staphylococcus aureus* was the most commonly isolated organism. Addition silicone impressions exhibited lower microbial loads than alginate.

Conclusion: Immersion in 2% glutaraldehyde or 0.5% sodium hypochlorite for 10 minutes effectively disinfects alginate and addition silicone impressions without compromising dimensional accuracy. These findings support the inclusion of time-controlled immersion disinfection protocols in clinical and laboratory practice to prevent cross-infection.

Keywords: Dental impression disinfection, Alginate, Addition silicone, Dimensional stability, Antimicrobial efficacy.

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1. Introduction

The oral cavity harbors nearly 1,000 species of microorganisms, collectively numbering in a magnitude of billions.[1] These microbes are a cause of concern for cross-infection to the dental professionals who perform treatment

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procedures in the oral cavity of the patients. Several species of microorganisms including the *Staphylococcus aureus*, *Pseudomonas*, *Streptococcus*, *Micrococcus*, *Bacillus* and *Candida Albicans* can thrive outside the oral environment even when not in the oral environment or in contact with the oral fluids.[2] The risk of cross-infection is, thus, not only limited to the dental operators but also extends to the laboratory personnel who deal with materials derived from the oral cavity (for instance, pathological laboratories) or materials that have been in contact with the oral tissues (eg: prosthodontic laboratory technicians or dental mechanics).[3,4] Additionally, students working in dental colleges handle these materials themselves during the fabrication of dental prostheses putting them at a risk of cross-infection.

Dental impressions are one of the procedures wherein the impression material as well as the tray are contaminated by the patient's saliva and/or blood, potentially serving as a source of cross-infection during laboratory procedures.[5] Therefore, the Centre of Disease Control and Prevention (CDC) guidelines dictate that all the dental impressions should be rinsed under running water, cleaned and disinfected using an effective hospital disinfectant before handling in the laboratory for infection control in the dental health care set up.6 Various disinfectants such as sodium hypochlorite, glutaraldehyde, and iodophors are available for disinfection of the dental impressions by either immersion or spraying method.[7] It is also a requisite that these disinfectants do not physically or chemically distort the obtained impression, implying that the dimensional accuracy and surface topography of the impression need to be preserved.

While the impending hazard of cross-contamination through dental impressions and the available means to counter it are common knowledge to dental students and professionals, their use is not a part of the standard operating protocol of many dental institutions and clinics. A cause of concern was highlighted by Marya CM *et al*. that about 75% of dental professionals across 60 dental colleges of India simply washed the impressions under running water without disinfecting them.[8] The question of whether merely washing with water is sufficient to eliminate the risk of cross-infection or whether the use of disinfectants is mandatory needs to be addressed. Evidence in this regard would dictate laboratory protocols in Indian dental institutions and motivate dental students and professionals to follow them.

In this context, the present study was conducted to assess the antimicrobial efficacy of two chemical disinfectants on Irreversible hydrocolloid (Alginate) and Addition Silicone impression material and also to assess the effect of these on the dimensional stability of the two impression materials.

2. Materials and Methods

The present ex vivo study was conducted in Bharati Vidyapeeth Dental College and Hospital, Kharghar, Navi Mumbai over a period of two years from August 2021 to August 2023. The study was conducted in accordance with the modified Helsinki declaration for good practices in research, and the protocol was approved by the institutional ethical review board (Ref ID: IEC320082021, dated 13/08/2021). Participants for the study were recruited from the patients visiting the institutional outpatient Department of Prosthodontics. Informed consent was obtained from all participants.

2.1. Patient selection

Systemically healthy patients aged 18 to 60 years were included in the study. A detailed case history was recorded, followed by clinical examination by trained investigators (MJ and MG). Patients with good systemic health and healthy periodontal tissues were included in the study. Patients wearing removable or fixed prostheses or those with a history of orthodontic treatment or periodontal surgery within the past six months were excluded. Smokers, tobacco chewers, and pregnant or lactating females were also excluded.

The gingival index was scored according to Loe and Silness criteria, and patients with scores 0 or 1, indicative of good gingival health, were included.[9] In cases where a lesion was present in the oral cavity or the periodontal health was compromised due to any pathology, the patients were excluded. Individuals who had received antibiotics, antifungals, or any form of immunosuppressive or chemotherapy for the past 6 months were also excluded from the study.

2.2. Sample size calculation:

The estimation of the sample size for the present study was based on the reported data for the mean values for log(CFU/cm2) with Addition Silicone and Alginate in a previous study by Demajo *et al*.[10] The sample size was calculated using one-way ANOVA (F test for group effect) H0: delta = 0 versus H1: delta!= 0. Keeping the α error as 0.05 and the power (1-β) as 0.9, the total sample size inclusive of six subgroups was estimated to be 48 for each outcome (microbial analysis and dimensional stability) respectively. However, to make the data more robust, it was decided that 15 samples would be included per subgroup, making the total sample size as 180.

2.3. Preliminary preparation

A preliminary impression of the maxillary arch was obtained using Irreversible Hydrocolloid (Alginate) impression material (Zhermack Tropicalgin, Badia Polesine (RO), Italy) with a perforated stock metal tray. The cast was immediately poured using Die stone (Type IV Gypsum) on the impression material. Four points were marked on the cast, which included the outermost points on the buccal height of contour of the first molars and canines bilaterally. These points were labelled A, B, C, and D, as indicated in the diagram (**Figure 1**). The distance between these points was measured using Vernier Calipers and considered as a comparative standard

for assessment of the dimensional stability of the disinfected impressions. The patient was recalled after seven days.

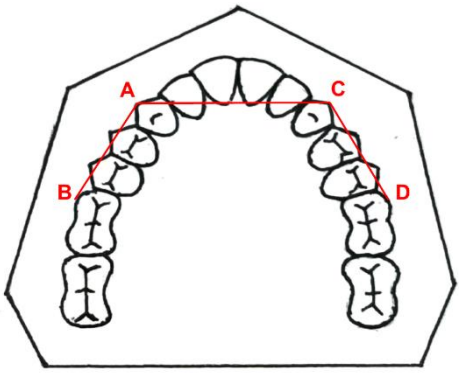

Figure 1: Distance measured between fixed points marked on the cast for assessment of dimensional stability

2.4. Preparation of the custom tray:

The outline of the custom tray was marked using an eosin pencil on the prepared die stone cast. Two layers of modelling wax were adapted to get a total of 4 mm space for the impression material. Four wax stoppers were made, two near the mesiobuccal cusp tip of the first molars and near the cusp tip of canines bilaterally. The light-cure acrylic tray material was adapted over the stone cast. A small vertical handle of the same material, of dimensions 25 mm x 12 mm, was attached to the edge of the labial border of the tray for easy placement and removal of the tray. Escape holes 12.5 mm apart were made with a tungsten carbide round bur, allowing the excess impression material to flow. Six such trays were made for each patient for the six addition silicone impressions.

2.5. Impression and disinfection procedures

In their second visit, twelve impressions were obtained from each patient, comprising six impressions each of a) Irreversible Hydrocolloid (Alginate) impression material (Zhermack Tropicalgin, Badia Polesine (RO), Italy) and b) Addition Silicone impression material (Aquasil, Dentsply), respectively. Stock trays of suitable size were used to obtain the alginate impression, while custom trays with Universal Tray Adhesive (Medicept Dental, Harrow, United Kingdom) were used to obtain the latter. The impressions were taken alternatively for both materials at an interval of 15 minutes between each. The Loe and Silness Gingival index was re-verified each time before obtaining the impression. All impressions were obtained by fixed trained operators (AR and SM) while the other laboratory procedures were performed by other two investigators. (MJ and MG)

All the impressions were washed in sterile water for 20 seconds. Following this, the impressions were assigned to Group A (Control), Group B (Glutaraldehyde), and Group C (Sodium Hypochlorite). The impressions in Group B and Group C were immersed in 2% Gluteraldehyde (3M Glutarex, India) and 0.5% Sodium Hypochlorite (Prime Dental, India), respectively, for 10 minutes. Six impressions (three alginate and three addition silicone for each patient) were immediately poured with die stone following the respective disinfection method to obtain a dental cast for assessment of dimensional stability. The dimensional stability was measured using the same method as described above for the cast prepared in the first appointment and was compared to the dimensions of the latter. The remaining six impressions were collected in a sterile pouch/container containing phosphate-buffered solution (PBS) and sent immediately to the institutional microbiological laboratory immediately after disinfection. The overall study procedure is delineated in **Figure 2**.

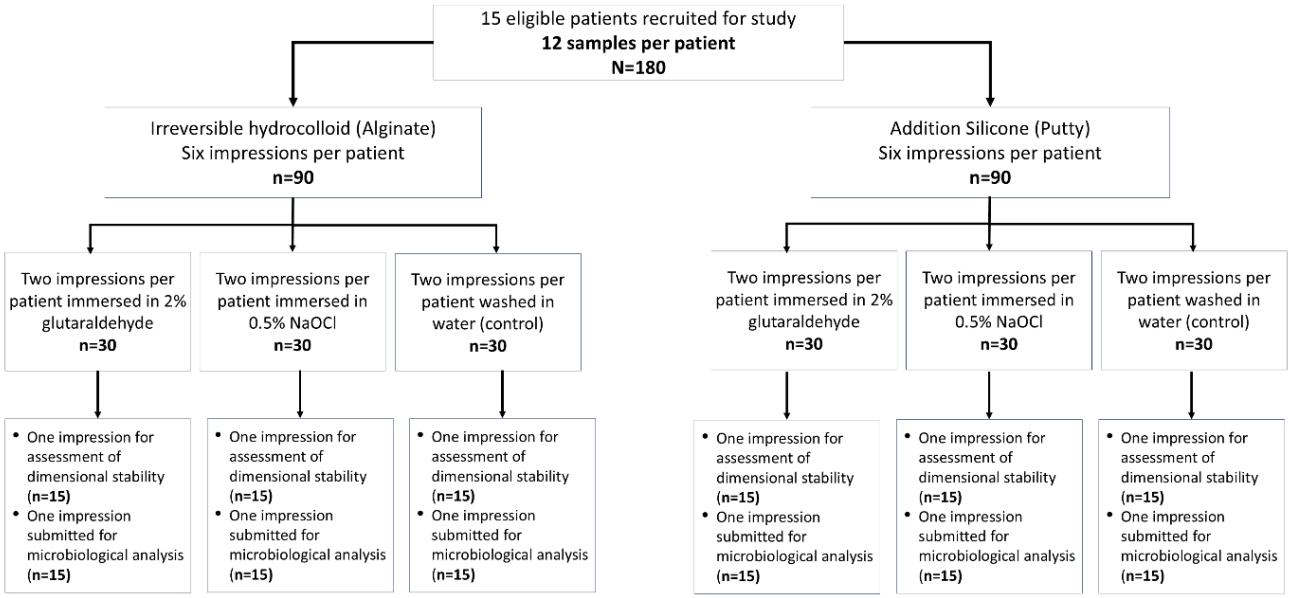

Figure 2: Flow diagram indicating the study process from patient inclusion to division of impressions into subgroups

2.6. Microbiological procedure

Samples from the PBS were inoculated on Nutrient agar, Blood agar, and MacConkey agar plates. These plates were incubated at 37°C for 24 hours. The colonies that appeared on the nutrient agar were counted using a colony counter to assess the number of bacterial cells as colony-forming units (CFU). The colonies that grew on Blood agar and MacConkey agar were subjected to biochemical tests (urease, citrate, and indole ring tests) for identification of bacterial strains (**Figure 3**). Likewise, Impressions without disinfection (control group) were subjected to the same process to draw a comparison between the study groups.

2.7. Statistical analysis

All data was entered into a Microsoft Office Excel (Office version 365) spreadsheet and checked for errors and discrepancies. Data analysis was done using the Windows-based 'MedCalc Statistical Software' version 19.0.6 (MedCalc Software bvba, Ostend, Belgium; http://www.medcalc.rorg; 2020). Data for the dimensional stability was expressed as means with standard deviation, whereas data for antimicrobial activity was expressed as log_{10}CFU with 95% C.I. The former were analyzed using parametric tests, and the latter by non-parametric tests.

The comparison of the three groups (2% Glutaraldehyde and 0.5% Sodium Hypochlorite, and Control) was done for dimensional stability and antimicrobial activity in the different impression materials using a two-way analysis of variance (ANOVA), with group as one factor and impression material as the second factor. Post-hoc individual pairwise comparisons were done using Bonferroni's test. Log CFU/cm2 were calculated and analysed for differences between the six groups using the Kruskal-Wallis test. All testing was done using two-sided tests with alpha 0.05.

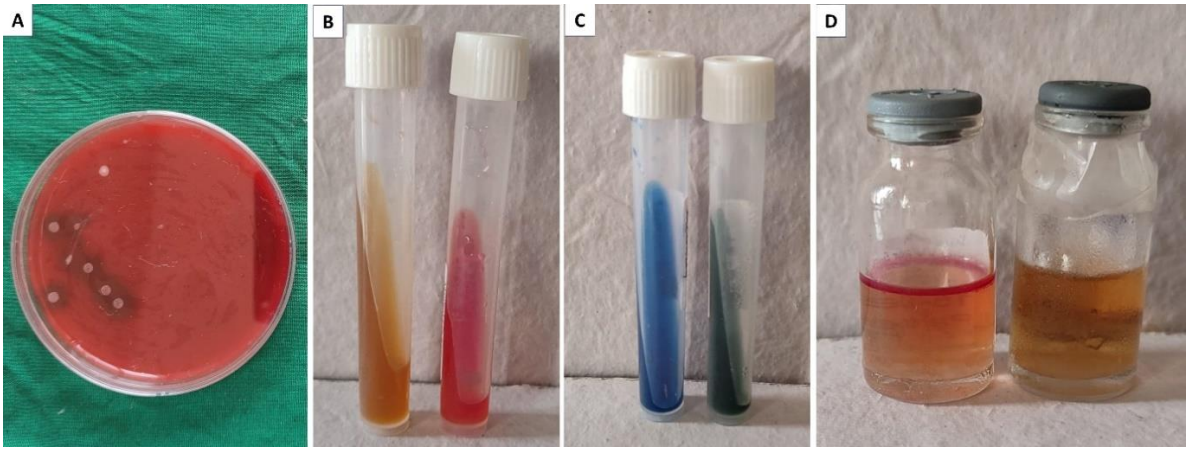

Figure 3: A): Hemolysis around microbial colonies on blood agar, **B):** Urease test, **C)**: Citrate test, and **D)**: Indole ring test

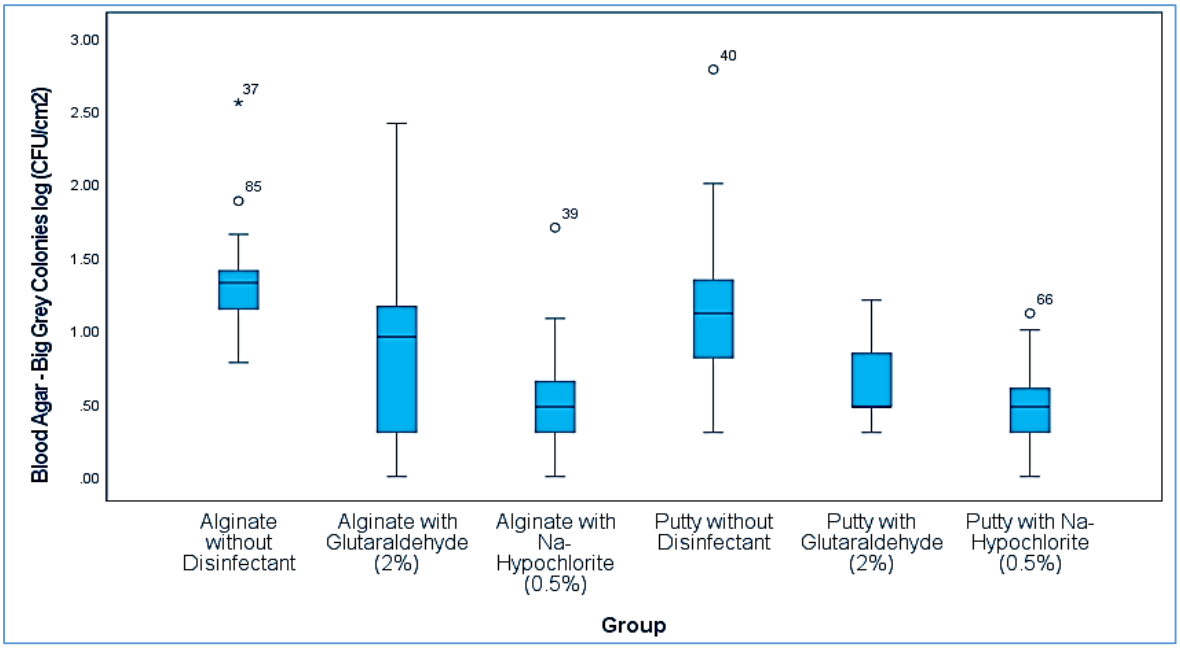

Figure 4: Box-and-whiskers plot for Log CFU/cm2: Median with percentiles

3. Results

3.1. Dimensional stability

The mean distance of dimensional averages for each subgroup is collectively displayed in **Table 1**. It was observed that the measured dimensions for the control and glutaraldehyde groups were lower than the alginate impressions disinfected with sodium hypochlorite. On the contrary, putty impressions showed an order of control>putty>alginate in the measured distance between the fixed points. Nevertheless, application of the two-way ANOVA test revealed no statistical difference in the dimensional stability based on the material or the disinfectant used (**Table 2**).

To better understand the differences between the effects of different disinfectants on both respective materials, a post hoc test between each subgroup was performed. Findings confirmed that the disinfectants did not have any effect on the dimensional stability of either material (**Table 3**).

3.2. Antimicrobial efficacy

No significant difference was observed between the microbial colony counts of both materials and the control in the McConkey Agar medium. The log of total CFU is depicted in **Figure 4**. In the blood agar medium, small cream colonies and big grey colonies were observed. A statistically significant difference ($p<0.05$) was observed in the number of colonies across the subgroups, with a higher number of colonies in the impressions not immersed in any disinfectant. Lower microbial counts were observed in putty as compared to their corresponding subgroups of alginate, and in impressions of the same material disinfected by sodium hypochlorite as compared to glutaraldehyde (**Table 4**).

The majority of organisms identified on Gram staining (**Table 5**) were gram-positive cocci (about 73 to 87%), followed by gram-negative bacilli (6 to 26%). The individual bacterial species identified in each subgroup are listed in **Table 6**. *Staphylococcus aureus* was the most frequently identified strain in 73 to 87% of cases, corresponding to the frequency of gram-positive cocci identified during gram staining, thereby confirming the findings.

Table 1: Mean dimensional averages of the distance between the fixed points for all subgroups

Material	Mean of distance between the fixed points (mm)	Mean	SD	N
Alginate	No disinfectant (Control)	24.45	1.967	15
	Glutaraldehyde (2%)	24.53	1.823	15
	Sodium Hypochlorite (0.5%)	25.03	2.090	15
Putty	No disinfectant (Control)	24.87	2.207	15
	Glutaraldehyde (2%)	24.79	2.093	15
	Sodium Hypochlorite (0.5%)	24.83	2.278	15
Total	No disinfectant (Control)	24.66	2.066	30
	Glutaraldehyde (2%)	24.66	1.933	30
	Sodium Hypochlorite (0.5%)	24.93	2.151	30

Table 2: Two-way ANOVA test applied to ascertain the significance of the effect of impression material and the disinfectant on the dimensional stability

Source	Sum of Squares	df	Mean Square	'F'	'p'
Corrected Model	3.66	5	0.73	0.169	0.973
Intercept	55135.08	1	55135.08	12721.006	0.000
Material	0.60	1	0.60	0.139	0.710
Disinfectant	1.49	2	0.75	0.172	0.842
Material * Disinfectant	1.56	2	0.78	0.180	0.836
Error	364.07	84	4.33		
Total	55502.81	90			
Corrected Total	367.73	89			
$R^2 = 0.010$ (Adjusted $R^2 = -0.049$)					

Table 3: Post-hoc test for pairwise comparison of disinfectant on the dimensional stability

(I) Disinfectant	(J) Disinfectant	Mean Difference (I-J)	SE	'p'	95% C.I. for diff.	
					Lower	*Upper*
No disinfectant (Control)	Glutaraldehyde (2%)	0.004	0.538	0.995	-1.065	1.073
	Sodium Hypochlorite (0.5%)	-0.271	0.538	0.615	-1.340	0.798
Glutaraldehyde (2%)	No disinfectant (Control)	-0.004	0.538	0.995	-1.073	1.065
	Sodium Hypochlorite (0.5%)	-0.275	0.538	0.610	-1.344	0.794
Sodium Hypochlorite (0.5%)	No disinfectant (Control)	0.271	0.538	0.615	-0.798	1.340
	Glutaraldehyde (2%)	0.275	0.538	0.610	-0.794	1.344

Table 4: Microbial counts observed across different subgroups in CFU/cm2

	N	Mean	SD	Median	Percentile		Range	Kruskal-Wallis test	
				50^{th}	25^{th}	75^{th}		*H*	*p'*
McConkey Agar - Colonies								5.106	0.403
Alginate without Disinfectant	2	0.13	0.35	0	0	0	1		
Alginate with Glutaraldehyde (2%)	0	0.00	0.00	0	0	0	0		
Alginate with Sodium Hypochlorite (0.5%)	1	0.07	0.26	0	0	0	1		
Putty without Disinfectant	1	5.33	20.66	0	0	0	80		
Putty with Glutaraldehyde (2%)	0	0.00	0.00	0	0	0	0		
Putty with Sodium Hypochlorite (0.5%)	0	0.00	0.00	0	0	0	0		
Blood Agar - Small Cream colonies									
Alginate without Disinfectant	15	103.60	181.18	27	11	38	593	34.082	<0.001
Alginate with Glutaraldehyde (2%)	15	12.27	10.82	10	5	15	42		
Alginate with Sodium Hypochlorite (0.5%)	14	7.60	9.49	5	3	7	35		
Putty without Disinfectant	15	95.47	173.27	15	5	40	547		
Putty with Glutaraldehyde (2%)	15	6.00	5.10	4	2	10	16		
Putty with Sodium Hypochlorite (0.5%)	15	4.93	4.51	2	2	8	13		
Blood Agar - Big Grey colonies									
Alginate without Disinfectant	15	46.00	87.73	21	12	27	351	34.642	<0.001
Alginate with Glutaraldehyde (2%)	15	26.60	65.20	9	2	15	257		
Alginate with Sodium Hypochlorite (0.5%)	15	6.67	12.30	3	2	5	49		
Putty without Disinfectant	15	58.87	151.65	13	6	22	598		
Putty with Glutaraldehyde (2%)	15	5.67	4.95	3	3	8	14		
Putty with Sodium Hypochlorite (0.5%)	15	4.00	3.44	3	2	4	12		

Table 5: Organisms isolated on Gram staining of the colonies obtained after inoculation of samples in agar

	Gram +ve bacilli		Gram +ve cocci		Gram -ve cocci		Gram -ve bacilli		
	No.	%	No.	%	No.	%	No.	%	N
Alginate without Disinfectant	1	6.7%	12	80.0%	1	6.7%	1	6.7%	15
Alginate with Glutaraldehyde (2%)	0	0.0%	13	86.7%	0	0.0%	2	13.3%	15
Alginate with Sodium Hypochlorite (0.5%)	0	0.0%	13	86.7%	0	0.0%	2	13.3%	15
Putty without Disinfectant	0	0.0%	11	73.3%	0	0.0%	4	26.7%	15
Putty with Glutaraldehyde (2%)	0	0.0%	13	86.7%	0	0.0%	2	13.3%	15
Putty with Sodium Hypochlorite (0.5%)	0	0.0%	13	86.7%	0	0.0%	2	13.3%	15

Table 6: Bacterial species identified in agar inoculated with samples of different subgroups

	Bacillus species		*Citrobacter species*		*Klebsiella species*		*Staphylococcus aureus*		
	No.	%	No.	%	No.	%	No.	%	N
Alginate without Disinfectant	1	6.7%	1	6.7%	1	6.7%	12	80.0%	15
Alginate with Glutaraldehyde (2%)	2	13.3%	0	0.0%	0	0.0%	13	86.7%	15
Alginate with Sodium Hypochlorite (0.5%)	2	13.3%	0	0.0%	0	0.0%	13	86.7%	15
Putty without Disinfectant	4	26.7%	0	0.0%	0	0.0%	11	73.3%	15
Putty with Glutaraldehyde (2%)	2	13.3%	0	0.0%	0	0.0%	13	86.7%	15
Putty with Sodium Hypochlorite (0.5%)	2	13.3%	0	0.0%	0	0.0%	13	86.7%	15

4. Discussion

Infection control remains a cornerstone of clinical dental practice, especially in the context of procedures that involve direct patient contact and the use of materials prone to microbial contamination.[6] Dental impression materials have been shown to harbor a broad spectrum of microorganisms, including *Staphylococcus aureus*, *Pseudomonas*, *Streptococcus* spp., and *Candida albicans*.[7] The present ex vivo study was conducted to evaluate and compare the antimicrobial efficacy of 2% glutaraldehyde and 0.5% sodium hypochlorite and their impact on the dimensional stability of two commonly used dental impression materials, alginate and addition silicone, using an immersion disinfection protocol. The results demonstrated that both disinfectants were highly effective in reducing microbial contamination and did not compromise the dimensional accuracy of the impression materials when performed within appropriate time frames.

One of the key findings in this study was that dimensional stability remained unaltered after immersion in either 2% glutaraldehyde or 0.5% sodium hypochlorite for 10 minutes. No statistically significant differences were observed in any of the measured dimensions across the groups, confirming that both disinfectants are compatible with alginate and addition silicone impressions when used as per recommended protocols. The American Dental Association recommends that elastomeric impression materials should not exceed a 0.5% dimensional change post-disinfection.[11] This threshold that was respected in the present study, validating the safety of the selected immersion time. These findings are consistent with Demajo *et al*., who also employed an immersion method with glutaraldehyde and reported no distortion in alginate or silicone impressions.[10] Similarly, Rad FH *et al.* and AlZain demonstrated clinically acceptable levels of dimensional change with immersion in glutaraldehyde and sodium hypochlorite, reinforcing the stability of these materials under time-restricted immersion.[12,13] These studies collectively validate the immersion protocol used in the present study and support the technique for disinfection, provided the exposure time does not exceed critical thresholds. The absence of significant distortion in the current study may be attributed to the precise control of disinfection time and material handling.

In contrast, Nimonkar *et al*. reported that immersion in 2% glutaraldehyde caused dimensional changes in vinyl polysiloxane.[14] This discrepancy can likely be attributed to extended exposure durations or differences in the hydrophilic nature and polymer cross-linking of the material used. Ismail *et al*., in his study, had highlighted that immersion of alginate impressions beyond the 10-minute duration could lead to dimensional compromise.[15] Therefore, concerns have been raised over the effects of the immersion method on the dimensional stability of impression materials. To overcome the drawbacks associated with immersion, some studies advocate spraying techniques over immersion, particularly for alginate impressions, to minimize dimensional alteration. However, this may compromise the antimicrobial effectiveness of the disinfection procedure. Qiu *et al*. also noted that immersion has higher antimicrobial effectiveness than spraying, particularly when disinfectant contact with all impression surfaces is critical.[16]

Dapello-Zevallos *et al.,* in their systematic review, emphasized that short immersion durations (≤10 minutes) in glutaraldehyde or hypochlorite solutions are generally safe for both alginate and elastomeric materials.[7] Another recent systematic review by Qiu *et al*. corroborated that both 0.5–1% sodium hypochlorite and 2% glutaraldehyde were highly effective for disinfecting alginate and elastomeric impressions, particularly when immersion durations were restricted to 10 minutes.[16] Notably, in our study, the 10-minute immersion window was strictly adhered to, which may have prevented any water absorption or material distortion, especially in hydrocolloid impressions that are inherently susceptible to syneresis and imbibition. Moreover, impressions were poured immediately after disinfection to minimize any post-treatment dimensional alterations due to water absorption or syneresis, especially in the case of alginate.[17]

With respect to antimicrobial efficacy, impressions that were just washed in water and not subjected to any disinfection protocol harbored a significantly higher number of microbial colonies. Specifically, in blood agar media, the CFU counts in the non-disinfected group were found to be 7 to 10 times higher than those of the disinfected groups. This stark difference underscores the critical importance of including a disinfection step in the clinical workflow before impressions are transferred to laboratories. The absence of such practices, as highlighted by Marya *et al*., remains a concerning trend in dental institutions.[8] Our findings caution the dental practitioners and students against the routine practice of merely washing the impressions under water without disinfection before pouring the cast or sending them to the laboratory.

Between the two disinfectants tested, 0.5% sodium hypochlorite demonstrated slightly superior antimicrobial efficacy compared to 2% glutaraldehyde. This is in agreement with findings from Pal *et al*. and Amin *et al*., both of whom reported that sodium hypochlorite was more efficient in eliminating microbial contaminants from impressions than glutaraldehyde.[18,19] The superior performance of hypochlorite may be attributed to its strong oxidizing potential, which disrupts microbial cell membranes and induces misfolding of intracellular proteins.[20,21]

Interestingly, addition silicone impressions exhibited consistently lower microbial counts than alginate impressions across all groups, including the control. This trend may be explained by differences in the inherent hydrophilicity and surface porosity of the materials. Alginate, being a hydrocolloid, has a porous structure and hydrophilic nature that supports microbial adhesion and retention of saliva and blood components.22 In contrast, addition silicones possess a more hydrophobic surface, limiting microbial colonization and facilitating easier removal of contaminants during rinsing and disinfection.[23] These material properties likely contributed to the observed differences in microbial load.

A noteworthy microbiological observation in this study was that *Staphylococcus aureus* was the most commonly isolated organism, appearing in 73–87% of all samples. This finding is of clinical significance as *S. aureus* is a known opportunistic pathogen that can cause serious infections in immunocompromised individuals. Previous studies by Ganavadiya *et al*. and Egusa *et al.* similarly identified *S. aureus* as a predominant isolate from contaminated dental impressions.[24,25] The current study, therefore, emphasizes that any disinfecting agent used in dental settings must be effective against this organism. Both glutaraldehyde and sodium hypochlorite, as confirmed by this study, achieved substantial reduction or elimination of *S. aureus*, validating their suitability for clinical use.

In the present study, microbial analysis involved both quantitative (CFU counts) and qualitative (gram staining and strain identification) parameters, which strengthened the reliability of the results. However, limitations include the lack of evaluation of surface detail reproduction, wettability, and long-term dimensional changes, which could be explored in future studies to further optimize impression disinfection protocols. Future research may also compare the efficacy of different disinfectants, especially herbal or organic ones, and also compare different methods of disinfection such as immersion and spraying.

5. Conclusion

Within the limitations of this ex vivo study, it can be concluded that immersion disinfection using 2% glutaraldehyde and 0.5% sodium hypochlorite for 10 minutes is both microbiologically effective and dimensionally stable for irreversible hydrocolloid (alginate) and addition silicone impression materials. Both disinfectants significantly reduced microbial contamination, with sodium hypochlorite demonstrating slightly superior antimicrobial efficacy. Importantly, no significant dimensional changes were observed in any group, supporting the safe use of these agents for impression disinfection. These findings reinforce the need for strict disinfection protocols in clinical practice to prevent cross-contamination, particularly against common pathogens such as *Staphylococcus aureus*.

6. Source of Funding

None.

7. Conflict of Interest

The authors have no conflicts of interest to declare. The authors are solely responsible for the content and writing of the paper. The authors have no affiliation with or involvement in any organization or entity with any financial or non-financial interest in the subject matter or materials discussed in this manuscript.

8. Ethical Approval

This study was approved by Institute Ethical Approval committee with Protocol No.: IEC 320082021 Version No.: 001 Dated: 13/08/2021.

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