

Evaluation of compressive strength, color stability, and antimicrobial action of chitosan-modified temporary crowns

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ABSTRACT

This research was aimed at observing how antibacterial strength, colour stability, and compressive strength of chitosan modified PMMA compare to non modified PMMA [polymethyl methacrylate]. The study consisted of 2 groups - chitosan modified PMMA was the test group while unmodified PMMA was the control group. Twenty-four specimens were prepared for each group. Compressive strength was evaluated using the Universal testing machine. The antimicrobial action against streptococcus mutans and lactobacillus was evaluated using the disc diffusion method. A reflectance spectrophotometer was used to measure the baseline colour following the CIE L*a*b* scheme. Following these experiments, the specimens were submerged in coffee and distilled water solutions [$n = 8$] for 15 days each. Color stability was measured by comparing the coordinates obtained pre and post the ageing method. Independent t test used to examine data on colour change and compressive strength. [$\alpha = 0.05$] It was observed that the incorporation of chitosan into polymethylmethacrylate increases its compressive strength. This was statistically significant [$P = 0.00$]. Improved colour stability was also observed [$P = 0.000$]. Antimicrobial action against streptococcus mutans and lactobacillus was seen in the chitosan modified group. Chitosan incorporation provides a promising improvement in the properties of the polymethylmethacrylate however further research with invivo studies are required to come to a conclusion.

Key words: Antimicrobial, chitosan, color stability, compressive strength, dental restoration, innovation, nanoparticle, polymethyl methacrylate, temporary

INTRODUCTION

Temporary restorations are a critical component of prosthetic clinical steps that safeguard the remaining

tooth structure during dental operations.^[1] They serve an important role until the luting of the final restoration.^[2] They deliver strength, retention, and esthetics to the prepared teeth, all of which are critical for clinical success. Temporary restorations are most commonly manufactured using polymethyl methacrylate (PMMA), polyethyl methacrylate, and urethane dimethacrylate.^[3,4] The desirability of any particular provisional restorative material is influenced by a number of factors such as fracture toughness, marginal accuracy, color stability, wear

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resistance, tissue compatibility, simplicity of manipulation, and affordability.

In recent times, nanotechnology has become a field of extensive research. The size of nanoparticles ranges from 1 to 100 nm. Because of the reduction in dimension to an atomic level, they have unique features. Chitosan is a nitrogenous, white, hard, inelastic polysaccharide formed by partial deacetylation of chitin.^[5] Chitosan nanoparticles [ChNPs] integrate the features of chitosan along with the qualities of nanoparticles, such as the surface interface, compact size, and effects of its quantum size.^[6] Owing to its antibacterial characteristics, nontoxicity, and biodegradability, it has a wide range of applications.^[1] Because of the listed benefits, chitosan can be mixed with other biomaterials to enhance their biological and mechanical properties.^[7] The use of chitosan materials in the dental world such as the alteration of dentifrices or dental luting agents has previously displayed promising results.^[8] Any changes to the composition must enhance or at least keep the original material's esthetic qualities and surface characteristics from declining.^[9] The goal of this research is to see how additives such as chitosan affect the strength, color stability, and antibacterial action of PMMA-based temporary restorations. The null hypothesis was that the compressive strength, color stability, and antimicrobial activity of chitosan-modified PMMA would have no statistically different mean values compared to control PMMA. Our research and knowledge have resulted in high-quality publications from our team.^[10-35] This study will give knowledge about the material and its modification.

MATERIALS AND METHODS

Chitosan nanoparticle preparation

0.5 g of chitosan powder was mixed with 0.5 g of glacial acetic acid and 49 ml of distilled water. This solution was kept on a magnetic stirrer for constant mixing, till a clear solution was formed. Then, 4–5 drops of sodium tripolyphosphate were

added. Characterization was done through transmission electron microscopy analysis [Figure 1].

Sample preparation

The ChNPs were directly incorporated into PMMA resin in the ratio of 1:2.5:1. For the antimicrobial test, ten specimens of each material measuring 1 cm × 1.5 cm were created, and samples of these materials were added in a metal mold, which was then coated with a polyester film and a glass slide, and pressed to eliminate extra materials and avoid the incorporation of air bubbles. The timer for the materials was set for a duration of 10 min. The specimens were kept at a temperature of 37°C and relative humidity of 100% for 24 h to guarantee full polymerization. After this period, the samples were polished on a polishing machine (APL-4) with 360-, 600-, and 1200-grit abrasive papers and completed with a solution containing diamond abrasives (6, 3, and 1 m) for a duration of 4 min each. Between and after the steps of polishing–finishing, the samples were rinsed with deionized water for 120 s using an ultrasonic cleaning device.

Color measurement

Color analysis of the specimens was carried out using the VITA Easyshade Advance digital spectrophotometer, following the (CIE L*a*b*) color scale at 10 a.m on a sunny day in sunlight when all other lights were switched off. All samples were analyzed on a single day.

Immersion in solution

Following the measurement, each specimen was immersed in containers containing coffee with a pH of 5.10 and distilled water (pH - 6.37) for 14 days at room temperature (37°C). Following the same process as before, new color measurements were taken after storage.

Color stability analysis

Following the new color measurements (3D color system), ΔE the aging treatments before and after were calculated

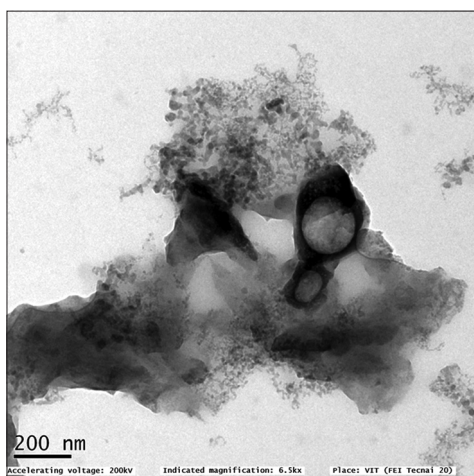


Figure 1: TEM image of chitosan nanoparticles. TEM: Transmission electron microscopy



Figure 2: Zone of inhibition seen against *Streptococcus mutans* in the sample modified by chitosan nanoparticles

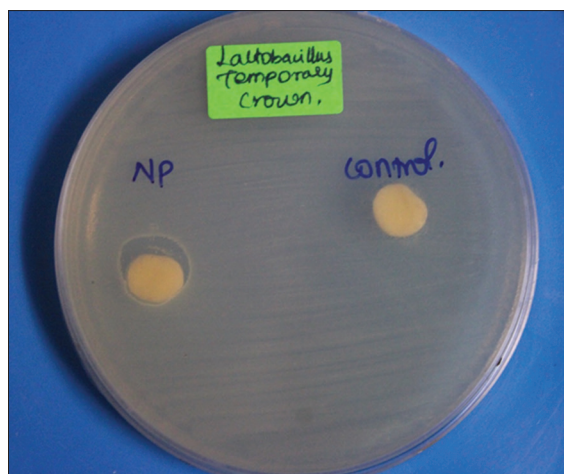


Figure 3: Zone of inhibition seen against *Lactobacillus* in the sample modified by chitosan nanoparticles

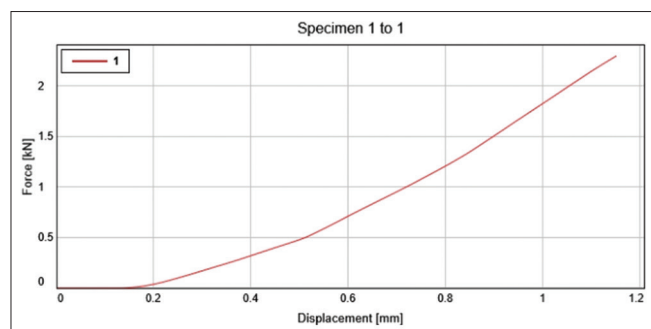


Figure 4: It depicts the displacement of the control group- sample 1 at a given force. Where the y-axis depicts the Force (kN) and x-axis depicts the displacement (mm)

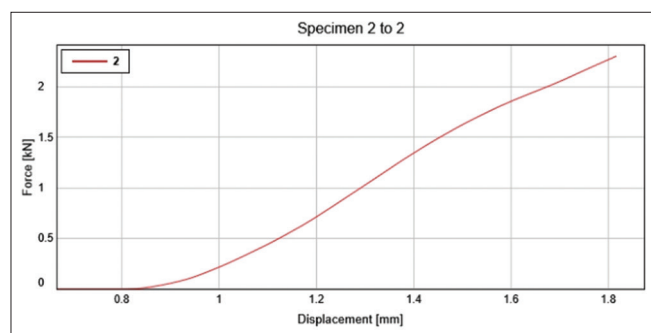


Figure 5: It depicts the displacement of the test group- sample 1 at a given force. Where the y-axis depicts the Force (kN) and x-axis depicts the displacement (mm)

$(\Delta E = [(\Delta L^*)^2 + (\Delta a^*)^2 + (\Delta b^*)^2])$. Depending on the material under study and the aging process, the mean values were determined.

Antimicrobial test

The antimicrobial activity tests of these samples were carried out using the disk diffusion method. *Streptococcus mutans* species and *Lactobacillus* species were taken and inoculated in the nutrient agar under room temperature.

Once the samples were added to the nutrient agar in their respective concentration, they were placed in the incubator at a temperature of about 37°C for 24 h. After the incubation period, the samples were taken to observe for the zone of inhibition, which was then measured. The results were obtained and statistically analyzed [Figures 2 and 3].

Measuring compressive strength

The specimens were made following the aforesaid process and then placed on top of the Instron universal testing machine's platform. A load of 10 kN load cells was applied (crosshead speed of 0.75 mm/min). The force that the sample could withstand before deformation started was measured in Newton and translated to MPa with the help of testing machine software [Figures 4 and 5].

Statistical tests

Descriptive statistical analysis was used to analyze the color stability and compressive strength of the chitosan-modified sample. The data were entered in the SPSS software (IBM Corp. Released 2015. IBM SPSS Statistics for Windows, Version 23.0. Armonk, NY:IBM Corp.). An independent *t*-test was performed, and results were obtained in the form of tables and graphs.

RESULTS

Color stability

The result in Table 1 depicts that there was not any difference observed in the material poststorage in the distilled water solution ($P > 0.05$). However, the post being stored in the coffee solution, the material modified with chitosan displayed greater color stability when in comparison to the control group, both on day 1 and day 7. The results were statistically significant ($P = 0.00$).

Compressive strength

The compressive strength of the chitosan-modified group was greater than the control [Table 2]. The results were statistically significant ($P = 0.00$).

Antimicrobial action

It was observed that the chitosan-modified group had an antimicrobial action against both *Streptococcus* and *Lactobacillus*. It formed a zone of inhibition against both bacteria, whereas the test group did not [Table 3].

DISCUSSION

Provisional restorations greatly affect the success of the ongoing treatment. For a successful treatment, it is essential that they have adequate strength, good color stability to maintain esthetics, and prevent bacterial colonization.

Bacterial colonization on provisional prosthetic materials is higher than that on permanent prosthetic materials

Table 1: Color stability of chitosan-modified sample is greater in coffee solution on both day 1 and day 7 when compared to the unmodified sample

Solution	Groups	n	Mean	SD	P
Distilled water (Day 1)	Control	10	0.1600	0.01491	0.13
	Chitosan	10	0.1510	0.00994	
Coffee (Day 1)	Control	10	2.1180	0.02821	0.00
	Chitosan	10	1.6640	0.02011	
Distilled water (Day 15)	Control	10	0.2600	0.01886	0.161
	Chitosan	10	0.2490	0.01449	
Coffee (Day 15)	Control	10	3.0500	0.15811	0.00
	Chitosan	10	2.5280	0.01398	

This result is statistically significant ($P < 0.05$). SD: Standard deviation

Table 2: The compressive strength of the chitosan-modified sample was greater than the control group

	Group	n	Mean	SD	P
Maximum force	Control	10	2297.1840	1.44411	0.009
	Chitosan	10	2298.6280	0.56478	
Compressive strength	Control	10	29.2380	0.01751	0.00
	Chitosan	10	29.2680	0.01317	

SD: Standard deviation

Table 3: Depicting the zone of inhibition formed

Group	Zone of inhibition (mm)	
	<i>Streptococcus mutans</i>	<i>Lactobacillus species</i>
Chitosan-modified PMMA	14	13
Control group	0	0

A zone of inhibition is observed against both *Streptococcus mutans* and *Lactobacillus* in the chitosan-modified group. No zone of inhibition is formed in the control group. PMMA: Polymethyl methacrylate

due to the high surface roughness and low marginal adaptation of provisional prosthetic materials.^[36] Since polymethylmethacrylate crowns are porous, bacteria can colonize them. One way to encounter this issue was to cover up the PMMA surface with nanoparticle-based antimicrobial agents such as chitosan, however, the optimum solution is that the nanoparticles be contained in the polymer matrix, so that their release operates at the stage of biofilm formation. In the current study, it can be observed that the addition of ChNPs increased the antimicrobial action of the temporary crown on the tested microorganisms *S. mutans* and *Lactobacillus*. Chitosan has a bacteriostatic or bactericidal and anti-adhesion effect and can reduce biofilm formation. It also has higher penetration power than conventional antimicrobial agents due to its micro size.

The interaction of cationic chitosan with anionic cell surfaces increases membrane permeability and causes cellular material leakage, which could be the reason for chitosan's antibacterial mechanism. Chitosan may also interfere with the synthesis of mRNA and the embedding of proteins.^[2]

During the function, the provisional fixed partial dentures are subjected to a variety of compressive, tensile, and shear stresses.^[3,4] Accordingly, temporary crown material must have sufficient compressive strength to resist fracture to extend its life.^[5] Hence, it is essential to improve its mechanical properties to achieve more crack-resistant restorations. In the current study, a statistically significant increase was seen in the compressive strength of temporary PMMA crowns postaddition of chitosan. The highly crystalline structure of chitosan along with its presence within the resin matrix most likely contributed to the increase in compressive strength. The fine dispersion of nanoparticles in the PMMA matrix may also have contributed to the increase in mechanical strength.^[6] This was, however, different from the results obtained by a study conducted by.^[7,8] In their study, they observed that the compressive strength of acrylic resin significantly decreased after the addition of 2% and 4% (w/w) ChNPs. They claimed that chitosan in acrylic resins acted as an impurity in the PMMA matrix, which reduced its flexural strength.^[9] According to^[9,10] chitosan may have a negative impact on polymerization conversion and result in an increase in the amount of residual monomer which acts as a plasticizer further decreasing compressive strength.

It is important that the inclusion of ChNP does not affect the esthetics of the restoration, hence color stability tests were performed. In the performed study, it was seen that the addition of chitosan increased the color stability of the temporary restorations in the sample with coffee on both days 1 and 7. The results were statistically significant. This could be due to the addition of methacrylate chitosan (low molecular weight) into the network of polymethyl methacrylate to produce stable materials. Similar findings were observed in a study conducted by.^[7] They discovered that the chitosan-modified group exhibited better color stability after storage in distilled water as well as red wine.

CONCLUSION

The following findings can be drawn within the limitations of the current study:

Antimicrobial efficacy increased in the sample modified by ChNP against both *S. mutans* and *Lactobacillus*. The chitosan-modified sample demonstrated higher color stability and greater compressive strength than the nonmodified sample. Although the modification of PMMA with ChNP seems to be a promising temporary restorative material with good antimicrobial, color stability, and compressive strength properties, further studies with greater sample sizes are required to come to conclusive results.

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Conflicts of interest

There are no conflicts of interest.

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