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Adhesion Of Microflora And The Role Of Denitrifies In Colour Stability On Provisional Crowns: An In-Vitro Study

Research Article

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Abstract

Background: Provisional crowns have an essential role in dental practice. These materials are used in the fabrication of temporary restorations before the permanent prosthesis is placed.
Aim: The aim of this study was to evaluate the adhesion of microflora and the role of denitrifies in colour stability on provisional crowns.
Materials and Methods: Four types of provisional materials based on fabrication technique namely Protemp, Tooth coloured cold cure acrylic, Heat cure acrylic and CAD-CAM milled acrylic were considered for the study. 9 specimens (10mm X 10mm) of each material were fabricated (n=36). Adhesion of microflora was tested in 3 specimens of each material by incubating at 370c with S.mutans (n=12). Bacterial adherence on surfaces was assessed and counting of CFU was done. 6 specimens of each material (n=24) were used and the colour stability was evaluated after 21,000 cycles of brushing using tooth brush simulator and data was recorded for both pre-operative and post-operative ΔE values using a digital spectrophotometer. The obtained data was entered in excel spreadsheet and One way ANOVA was done using SPSS software version 23.0.
Results: The microbial adhesion was least in heat cure acrylic, followed by CAD-CAM milled acrylic, Protemp and tooth coloured cold cure acrylic ($p>0.05$).The colour stability of the was highest in heat cure acrylic , followed by CAD-CAM acrylic, tooth coloured cold cure acrylic and Protemp material ($p<0.05$).
Conclusion: From the results obtained, it can be concluded that heat cure acrylic showed best colour stability followed by CAD CAM acrylic and the least microbial adhesion was found in heat cure acrylic followed by CAD CAM acrylic. Further research required to come up with a material that serves good properties in both aspects.

Keywords: Colour Stability; Digital Spectrophotometer; Microbial Adhesion; Provisional Crowns; Toothbrush Simulator.

Introduction

Temporization materials are temporary crown and bridge materials. They are essential in the practice of dentistry [1]. It is a provisional prosthesis before the permanent prosthesis is placed. This property favours the dentists or lab technicians because they get the time they need to create a more proper permanent restoration without sacrificing patient's comfort and dental function [2]. In complex and advanced cases, temporization could serve the purpose of testing function, phonetics and aesthetics. This information can then be transferred to the laboratory for fabrication of the final or permanent prosthesis or restoration [3]. This acquires increased patient acceptance and satisfaction and acts as an important tool for the psychological management of the patient until the permanent restoration is fabricated [4, 5]. Typically, the use of these appliances is limited to several weeks. However, the effectiveness of these restorations will vary depending upon their application. For example, in implant dentistry, the temporary prosthesis may function for a longer waiting period before the permanent prosthesis is placed. On the other hand, the prosthesis in high-stress sites can throw up additional challenges even for short-term use. For this reason, a temporization material should have good environmental, chemical, mechanical and most importantly aesthetic stability in the oral environment [6]. The PMMA, Bis-GMA, bis-acryl composite resin, urethane dimethacrylate, polyethene methacrylate, polyvinyl methacrylate, etc are

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the materials that are currently being used for the fabrication of temporary prosthesis [7].

Colour stability of temporization materials is the ability of the materials to retain their colour in the oral environment. It is compromised by the daily intake of food and drinks like tea, turmeric, coffee, etc [8]. The present study deals with 4 different types of temporization materials namely Protemp, Tooth coloured cold cure acrylic, heat cure acrylic and CAD/CAM material. All these materials are polymethyl methacrylate (PMMA) based. An exception is Protemp, which is a bis-acryl composite resin-based. Protemp is a trend dental material, available in automix air-tight syringes and is used for virtually all interim restorations including crowns, veneers, inlays and Onlays with good edge-strength, least marginal discrepancies [9, 10]. Tooth coloured cold and heat cure materials are acrylic resins composed of PMMA. They are available in powder (polymer) and liquid (monomer) form and are used in the fabrication of dentures, splints and many other intraoral applications [11]. CAD/CAM was first introduced in dentistry in the 1970s. CAD/CAM technology has emerged recently in dentistry and fabricates materials with high precision by permitting shaping, preventing rotation of tooth from normal position and providing better Strength [12-14].

A toothbrush simulator is a mechanical simulator capable of running a programmable three-dimensional brushing pattern [15]. It's very useful to check the efficacy of a toothbrush [16], flexibility, abrasion and colour stability of restorative materials and prosthesis.

The hypothesis behind the study was mainly to determine the fact that the coloured dentrifrice can cause staining of provisional crowns which will affect the colour stability of the prosthesis, hence coloured dentrifice were used in the present study.

When provisional restoration is subjected to bacterial contamination, it may lead to the failure of the restoration [17]. There are three factors that lead to bacterial contamination and thus affect the success of a restoration. They include i) when the provisional prosthetic restorations are worn for prolonged durations, they will allow the formation of colonies of bacteria on their surface. ii) Another major factor is the species of bacteria. Streptococcus is one of the most common, early colonising bacteria and is known to be the primary pathogenesis of tooth caries. iii) One another factor is the organic and inorganic composition of the materials which contributes to microbial adhesion.

An ideal provisional restoration should have polished surfaces, tissue-friendly margins and contours to avoid irritation and to reduce the potential for staining. It should discourage the development of plaque and attachment of oral bacteria onto its surface [18, 19]. Considering the above parameters, this present study aimed to determine the adhesion of microflora and the role of denitrifies in colour stability on provisional crowns, hence four trending and commonly used temporary crown materials were evaluated and compared.

Materials and Methods

Specimen preparation

Four different types of temporization materials namely, protemp, tooth coloured cold cure acrylic, heat cure acrylic and CAD/CAM milled acrylic were taken and made into square shaped acrylic specimens. 10mmx10mmx2mm dimension design STL (Standard Tessellation Language) file was created in the software and it was milled using milling machine (MES-I core) for CAD CAM acrylic specimens. Same dimension was used to make index in dental flasks on plaster, following the dewaxing procedure the heat cure specimens were fabricated. Another flask for plaster mould was made to prepare tooth colour acrylic and protemp specimens. Total of 36 specimens of 2mm thickness were prepared of which 24 specimen were used for colour stability and 12 were used for microbial adhesion.

Test for microbial adhesion

Three fabricated specimens from each group were disinfected using cold sterilization to prevent contamination. Artificial saliva was taken in a sterile container and to it, 1% glucose was added. A bacterial suspension of Streptococcus mutans was added to the container. The slabs were immersed in the solution and the container was incubated for 3-4 hours at room temperature. After the desired duration, the container was removed and the specimens were rinsed by flushing saline. After rinsing, each specimen was put in a separate sterile container into which 5ml of saline was added. After some time, 50 µL was collected from each container and transferred to Petri plates containing TSA media. The Petri plates were then incubated overnight for bacterial culture and the bacterial count was checked manually the next morning.

Test for colour stability

6 specimens from each group were mounted on dental stone and placed in a toothbrush simulator (Figure 1A). Unused soft full head multi labelled tooth brushes (colgate) were placed on the tooth brush simulator. The samples were subjected to 21,000 cycles of brushing consisting of horizontal, vertical and zig-zag patterns. The coloured dentifrice used was Dabur red (Dabur Laal). The colour stability was checked using VITA Easy shade Advanced 4th generation digital spectrophotometer by recording and comparing the preoperative (before brushing) and post-operative (after 21,000 cycles of brushing) ΔE values (Figure 1B).

Statistical analysis

The obtained results were recorded and the one-way analysis of variance (ANOVA) test was performed using SPSS software version 23.0.

Results

The colour stability and microbial adhesion of different temporary crown materials was evaluated using One way ANOVA test wherein the discrepancies or differences between the preoperative and post-operative ΔE values were compared and the P-value was found (Table1).

The mean microbial count of Protemp material is 54.67 X104, Tooth coloured cold cure acrylic 62.00 X104, Heat cure acrylic 36.00 X104and CAD/CAM acrylic 50.33 X104 which overall showed least microbial adhesion in heat cure acrylic with the P value 0.350 ($p > 0.05$) which is statistically not significant (Figure2). The mean count of colour stability of Heat cure acrylic

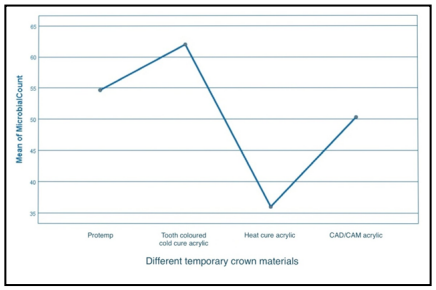
Table 1. Comparison of four different temporization materials based on microbial adhesion and colour stability (Heat cure acrylic, CAD/CAM acrylic material, Protemp, Tooth coloured cold cure acrylic).

Colour stability	Group	Mean	SD	Std. Error	95% CI	95% CI	F value	P-value
	Heat cure acrylic	0.267	0.057	0.033	0.123	0.41	6577	0.001*
	CAD/CAM acrylic	0.7	0	0	0.7	0.7		
	Protemp	2.9	0	0	2.9	2.9		
	Tooth coloured cold cure acrylic	2.7	0	0	2.7	2.7		
	Heat cure acrylic	36	16.46	9.5	4.89	76.89	1.263	0.35
	CAD/CAM acrylic	50.33	9.45	5.45	26.85	73.81		
	Protemp	54.67	18.58	10.72	8.5	100.83		
	Tooth coloured cold cure acrylic	62	20.8	12.01	10.31	113.69		

P-value derived from one way ANOVA test (*Significant at P<0.05)

Figure 1: A) Temporization materials with dentifrice under the cycle of Toothbrush simulation. B) Digital measurement of color stability through spectrophotometer.

Figure 2: Showing the microbial adhesion of the temporization materials. Toothcolour cold cure acrylic shows maximum bacterial adhesion followed by protemp, CAM CAM acrylic and heat cure acrylic, there is no significant difference between groups (p>0.05)



0.267, CAD/CAM acrylic 0.700, Protemp 2.900 and Tooth coloured cold cure acrylic 2.700 showing highest colour stability in heat cure acrylic when compared to other materials with p value 0.001 which is statically significant (p<0.05). After 21,000 cycles of brushing, the colour stability of heat cure acrylic was the best with only a slight difference in the pre-operative and post-operative ΔE values. This was followed by, CAD/CAM acrylic material, Tooth coloured cold cure acrylic and the difference was highest in protemp material. It can be observed from Figure 2 that the microbial adhesion was least in heat cure acrylic which is then followed by CAD/CAM material, protemp and the adhesion was highest in tooth coloured cold cure acrylic. Hence the results proved that the microbial adhesion was least in heat cure acrylic, followed by CAD-CAM milled acrylic, Protemp and tooth coloured cold cure acrylic and the colour stability of the was highest in heat cure acrylic , followed by CAD-CAM acrylic, tooth coloured cold cure acrylic and Protemp material.

Discussion

Provisional restorations play a very important role in dentistry. Hence, it is necessary to build these restorations with an ideal material that makes the interim restoration favour and resemble the properties of the permanent prosthesis for time-being. The success of a permanent restoration depends majorly on the success of the provisional restoration. Thus, the provisional restoration should be capable of fulfilling the needs expected from a permanent prosthesis; especially normal function and aesthetics, while the permanent prosthesis is being fabricated. Colour stability of the provisional restorative material is very important for the esthetic success of the interim restoration. The adhesion of microbes present in the oral cavity or saliva to the surface of the restorative material has a negative impact on pulpal protection and success of the permanent restoration. There are many other factors that need to be considered, which include pulpal protection, occlusal function, abrasion resistance, strength, etc in addition to esthetics and microbial adhesion. Thus, an ideal restorative material should serve all these purposes.

The preoperative (before brushing) and postoperative ΔE values were recorded using a digital spectrophotometer in order to check the colour stability of these materials. The results obtained from the present study imply that heat cure acrylic exhibited excellent colour stability, with only a slight difference between the

preoperative and post-operative ΔE values. The material showing second-best colour stability in the present study was CAD/CAM acrylic with a slight difference comparing with heat cure acrylic. Protemp material showed the lowest colour stability, with a significantly greater difference between the preoperative and post-operative ΔE values. A number of factors can contribute to these results namely, the surface roughness of the material, the chemical composition of the material and its interaction with the constituents of the toothpaste, ability of the material to be finished and polished, role of toothpaste in staining the material, colour absorbing property of the material, etc. Regarding microbial adhesion results imply that the adherence of bacteria to the surface of the specimens of tooth coloured cold cure acrylic was very high, followed by Protemp and CAD/CAM acrylic. Microbial/bacterial adhesion to the surface of heat cure acrylic material samples was the least. These results also depend on a number of factors including surface irregularities, the medium used for polishing, Edward J Givens, et.al. in 2008 tested the colour stability and marginal fit of Protemp Garant, Protemp Integrity, and Luxatemp Solar, against SNAP, a polymethyl methacrylate control and concluded that Protemp Garant exhibited a clinically noticeable change in the shade after 1 week in staining solution, whereas the other materials didn't change its colour [20]. These results are in correlation to the results obtained in the present study.

Provisional prosthetic materials show variable colour stability under different conditions in the oral cavity. The colour of the same prosthetic material may be perceived differently, depending on the source of light and the effect of the environment in which it is used. The study by Koczorowski et.al. [21], the colour stability of Luxatemp, Structur2S.C.,Protemp II, Zhermacryl STC and Dentalon Plus materials was evaluated by immersing the samples of these materials in coffee, tea and dark fruit juice for 60 hours followed by evaluation by a spectrophotometric method. The results were obtained in artificial light (illuminant A) and daylight (illuminant D65) and compared. It was found that Structure material exhibited the greatest tendency to discolouration. Protemp II, Luxatemp and Zhermacryl materials showed moderate discolouration and the least tendency to discolouration was exhibited by DentalonPlus.Almohareb et.al. [7] tested the colour stability of three temporary restorative materials (Systemp C&B, 3M Protemp 4, and Telio CAD) in four solutions (Pepsi, coffee, tea, and distilled water) using the CIELAB system and a colourimeter and found that the highest colour stability was exhibited CAD/CAM material, followed by Protemp and Systemp C&B with was in line with our study also. It was stated that the material can be used for long-term restoration and this property could be attributed to the pre-polymerization, higher monomer conversion, and minimum preparation errors in CAD/CAM materials7.In a study done by Soles JO et.al., [22] samples of Jet Set-4 (Lang), Protemp Plus (3M ESPE), Luxatemp (DMG), Artbloc (Merz), Telio-CAD (Ivoclar), and Vita-CAD (Vita) were immersed in: distilled water, coffee, red wine, tea, coke and cranberry juice. Colour testing was done by a spectrophotometer at baseline, 24 hours, 4 and 8 weeks. Specimens in coffee and red wine for 8 weeks were exposed to tooth-brushing for three minutes and colour measurements were taken subsequently. Colour differences (ΔE) after treatments were calculated. It was found that CAD/CAM blocks showed a significantly lower colour change, at all durations, and after brushing than the traditional materials which was supporting our study.

In a study, Kuphasuk et.al. [23] evaluated the flexural strength and colour stability of 3 bis-acryl resins (LuxatempFluorescenceTM, Protemp 4TM and IntegrityTM) and a polymethyl methacrylate-based resin (UnifastTradTM) after different ageing conditions. It was found that colour differences (ΔE) of all bis-acryl resins were lesser than that of the PMMA-based resin, which was similar to the results obtained in the present study. Zortuk, M et.al.tested microbial adhesion of samples of bis-acryl composite resins including protemp, PMMA based resins and a light-polymerized composite resin and found that bis-acryl composite resins showed more susceptibility to bacterial adhesion than PMMA based resins [17]. The results obtained were similar to those obtained in the present study, wherein bis-acryl based protemp material shows high susceptibility to bacterial adhesion. In a study, Ozel GS et.al. [24] compared the adhesion of S. mutans on bis-acryl resin material and PMMA based material. It was observed that S. mutans adhesion was the highest in bis-acrylic resin material and least in the PMMA based material. It was explained that this could be attributed to the surface free energy, which has a strong effect on bacterial adhesion. The hydrophobicity of the PMMA group is higher than the bis-acrylate group, therefore, the S. mutans adhesion is lower to PMMA based materials .Therefore in the present study, microbial adhesion and colour stability of 4 trending and commonly used temporary crown materials showed that heat cure acrylic and CAD CAM acrylic shows excellent colour stability and also it shows good antimicrobial property, owing to lesser microbial adhesion to its surface when compared to the other materials. . Similar literature available where abrasion resistance was compared for the same group of material [25]. This paper will add another point for selection of temporary crown material in clinical practice.

Conclusion

The CAD CAM is the modern temporary approach of prosthesis by digital means. The planning of the prosthesis takes place virtually in the 3D software which helps in minimizing maximum errors. These CAD CAM temporary crowns are fast, precise and almost mimicking final prosthesis which can be used for long term temporization for the advanced treatment like implants and full mouth rehabilitation where development of soft tissue architecture is a predominant demand along with esthetic outcome and function. On the other hand the heat cure acrylic crown fabrication is an old, tedious and time consuming, manual technique which may associated with processing errors, hence CAD CAM fabricated acrylic crown is a fantastic alternative for current dentistry trending into digital era. Further research required to come up with a material that serves good properties in both aspects.

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