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## Effect of Staining Solutions on Color Stability of Acrylic Denture Base Resins - A Spectrophotometric Evaluation

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### ABSTRACT

The purpose of this study was to quantitatively evaluate the effect of staining solutions by spectrophotometric method on color stability of the commercially available acrylic denture base resins polymerized by different methods of cure, materials, and immersion periods. Commercially available heat cure resin (Travelon) and self cure resin (RR repair material) were selected for the study. In case of the heat cure resin, the samples were prepared by both fast cure and slow cure polymerization techniques. The samples were immersed in staining agents: Turmeric, Yoghurt, Coffee, Tea, Cocoa and Lime for a period of one day, seven days and 30 days of daily use. At the end of each staining period, the color values were measured using a reflectance spectrophotometer and the color difference was determined amongst all the groups based on CIE Lab system. Statistical analysis of data on 30 days showed highest degree of color change in case of turmeric followed by yoghurt, tea, coffee, lime and cocoa. The heat cure slow samples were also most color stable followed by the heat cure fast samples. The self cure samples were found to be most susceptible to the color changes. The material, method of cure, immersion periods and staining solutions were significant factors that affected color stability of acrylic denture base resins. Public wearing prosthesis should be aware of their dietary habits, if their prosthesis needs to be worn for a long period. The study also aids the dentist in selecting resins for denture construction and denture repair procedures based upon color stability under laboratory condition.

**Keywords:** Color stability, Acrylic denture base resins, Spectrophotometer, Staining solutions.

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## INTRODUCTION

It is often difficult to restore a pleasing smile to patients who are psychologically depressed of having partially dentulous or edentulous oral conditions. Prosthesis made of acrylic polymers is the best remedy to this problem. These acrylic denture base resins (DBR) though have a variety of advantages have to always pass the test of time when color stability is concerned. In other words, failure or success of any esthetic material mainly depends on color match and the color stability of material in long term use.

Color stability, the property of material to retain its color over a period of time, in a specified environment, is an important property of many materials used in dentistry [1]. Color stability is required characteristic of the denture base polymers specified by various national and international standards typically American Dental Association specification number 12 [2] and may provide important information on serviceability of prosthetic materials. The requirement of color and translucency of prosthesis should be maintained during processing and these resins should not be stained or change color in clinical use.

Changes in optical properties of the material in a service environment have been responsible for color change or color instability [3]. Optical properties plays an important role in color matching, since the transmitted light with the optical or color information of the background and surrounding environment will affect the appearance of the resin. Discoloration (staining) is one among the factor that affects the optical property. Clinical and *in vitro* studies [4] have implicated dietary compounds as major etiological factor in staining of acrylic materials leading to patient dissatisfaction, additional time and expense for replacement of the prosthesis. Though they were good amount of studies in literature [4] on tea, coffee used as staining agents, there are limited literature studies to correlate the effect of turmeric, yoghurt, lime, and cocoa on DBR. While a number of authors have reported on color changes and staining of provisional prosthodontic materials [5,6], soft lining materials[3,7-9], acrylic denture teeth materials [10], crown and fixed partial denture resins [11], there are dearth of reports in literature on color stability of acrylic denture base materials [12-14]. Hence, in the present study turmeric, yoghurt, lime, cocoa, tea, coffee was taken as test agents to study the color stability of acrylic denture base resins.

The hypothesis to be tested in this study was that stain ability of DBR materials was related to material type, method of cure, staining solutions, immersion periods and dietary habits of patients.

## MATERIALS AND METHODS

In this study, six commonly consumed beverages were used as staining agents and distilled water as control, to evaluate the effects of color stability on commercially available, heat activated resin and chemically activated (self cure) resin. Table-1 and Table- 2 lists the details of the resins and staining agents used in the present study. It is known that obtaining conditions of oral environment *in vitro* is not easy. To simulate *in vivo* conditions in this study,

six different staining solutions and distilled water (control) were used at a constant temperature of  $37\pm 1^{\circ}\text{C}$ . The quantities of staining ingredients to make the test solutions were prepared closer to the amount consumed per person at a time. This study is an indicator of cumulative effect of repeated short immersions of acrylic resins used for prosthesis fabrication, during prolonged service.

**Table-1. Materials used for the study**

Resin used	Code	Manufacturer	Batch No.
<b>Trevalon</b> (Heat cure Resin, Shade-pink)	Heat Cure Fast- HCF Heat Cure Slow- HCS	Dentsply India Pvt Ltd	T071226
<b>RR Repid Repair powder</b> (Self cure Resin, Shade-pink)	SC	Dentsply India Pvt Ltd	R080201
Universal denture liquid		Dentsply India Pvt Ltd	VL071217

**Table- 2. Staining agents used for the study**

Staining Agents	Code	Manufacturer	Batch No.	Proportion
<b>Turmeric</b>	T	MTR Food Ltd, Bangalore, India	803011736	1g/ltr boiling water, simmer for 5 min & filtered
<b>Coffee</b>	C	Nescafe Sunrise Premium, Nestle India Ltd, New Delhi, India	7230452RB	30g/ltr boiling water, simmer for 5 min & filtered
<b>Tea</b>	E	Brooke Bond Red Label Hindustan Unilever Ltd Mumbai, India	U2304	30g/ltr boiling water, simmer for 5 min & filtered
<b>Cocoa</b>	O	Cadbury Cocoa, Cadbury India Ltd, Mumbai, India.	901233000312	30g/ltr boiling water, simmer for 5 min & filtered
<b>Lime</b>	L	Home made	--	100ml/ltr of distilled water
<b>Yoghurt</b>	Y	Home made	--	½ ltr

### Preparation of test solutions

30g of tea, coffee and cocoa and one gram of turmeric were separately taken in one litre of distilled water and solution was prepared by boiling, followed by simmering for five min and filtered through a filter paper. In case of lime, 100ml of lime juice concentrate was dissolved in one litre of distilled water.

### Preparation of test specimens

Acrylic specimens of 10mm diameter and 2mm thickness were prepared by lost wax investment technique. Type III Gypsum was used as investment material. Heat cure specimens were prepared by compression molding technique. Two curing cycles were followed: Slow cure (HCS) [ $74^{\circ}\text{C}$  for 8 hours] and Fast cure (HCF) [ $74^{\circ}\text{C}$  for one hour followed by  $100^{\circ}\text{C}$  for two hours]. Self cure acrylic resins (SC) was mixed according to manufacturer's instruction, packed into mold space and cured at room temperature. A total of 105 samples (35 samples for each group of HCS, HCF, SC resin) were fabricated which were to be stained for 1, 7 and 30 days time

interval. All the specimens were given identification number on them to avoid manual bias while evaluating through spectrophotometer. The samples were polished with different grades of Silicon Carbide paper (80, 100, and 120) and buff polished with pumice slurry.

## Method

The resin samples were stored in distilled water for 24 hour before immersing into the test solutions [15]. After the baseline measurement, the samples were immersed for 8 hour of immersion period daily (simulating overnight soaking period) for 1 day, 7 days and 30 days. The solutions were stirred twice a day to reduce precipitation. The solutions were refreshed each day. The samples were then rinsed in distilled water for 8-10 dips and blot dried with a tissue paper. They were stored in a desiccator for the remaining period until they were placed in a viewing port for color measurement. The control samples were stored in water at room temperature, the water being changed every day.

## Measurement of color

Color changes ( $\Delta E$ ) were measured objectively by a reflectance spectrophotometer (Spectrolino, Gretag-Macbeth AC, Germany), to potentially eliminate the subjective errors of color assessment. To measure chromatic differences Standard Commission International de L' Eclairage (CIELab) color system was used. It quantifies color in terms of three coordinate values  $L^*$ ,  $a^*$  and  $b^*$ .  $L^*$  represents brightness or lightness (value),  $a^*$  and  $b^*$  serve as numeric, correlates both for hue and chroma on green- red axis and blue- yellow axis respectively. The magnitude of color difference perceived between two objects is calculated by formula  $\Delta E = (\Delta L^2 + \Delta b^2 + \Delta a^2)^{1/2}$ . Before each measurement session, the instrument was calibrated according to manufacturer instruction by using the supplied white calibration standard. Color was measured according to CIE Lab color scale. D50 standard illuminant from a tungsten lamp was used with a viewing angle of  $2^\circ$ . UV filter was positioned to 100% UV. Color measurements were made in three randomly selected areas for each specimen and average of three reading was recorded. The mean and standard deviation was calculated.

## Statistical analysis

In all, there were 105 samples that provided a total of 315 reading for statistical analysis. The most appropriate statistical analysis was done by one way analysis of variance (ANOVA) (comparison between type of material and staining agents) followed by Tukeys HSD test for post hoc analysis. Unpaired- t tests was used to examine whether significant color difference occurred during immersion at specified time interval.  $P < 0.05$  was considered as significant.

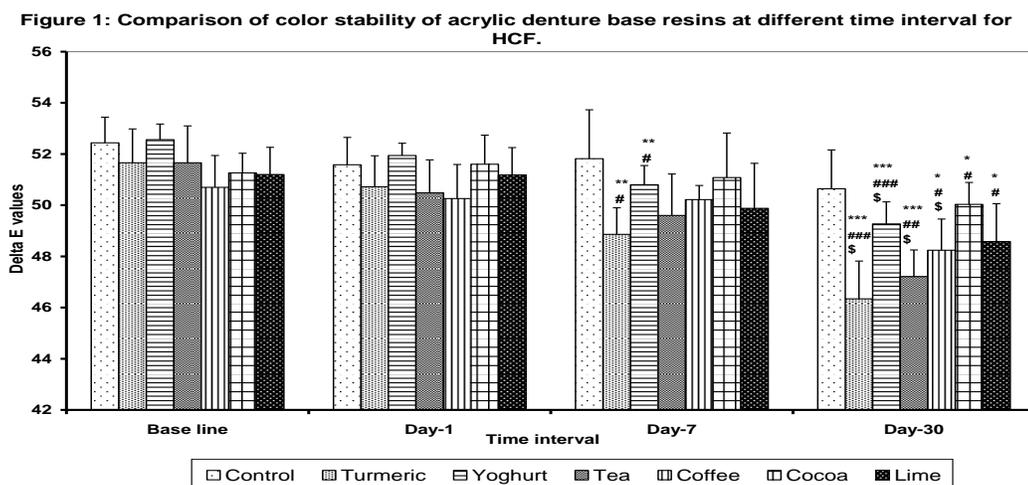
## RESULTS

Visual analysis showed significant color differences between material type, immersion period and staining agent. This was also corroborated by statistical analysis of data obtained from reflectance spectrophotometer. Immersion time was found to be a critical factor for color

stability of DBR. Unpaired- t test showed that as immersion time increased, color changes became more intense. The test showed statistical significant differences between means of baseline and day 30, baseline and day seven, day one and day 30, day seven and day 30 values. The color stability of the acrylic resins did not differ between base line values and day-1 measurement in any of the groups. The control samples showed color stability during entire experimental period (day one to day 30) in all the groups.

**Comparison at different time interval**

**(a) With HCF samples**

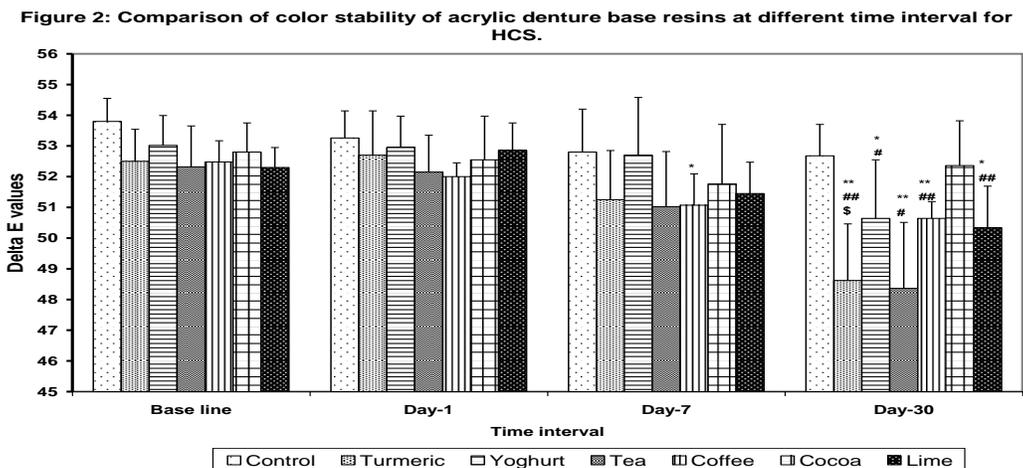


Comparison with base line values: \* = p<0.05, \*\* = p<0.01 and \*\*\* = p<0.001  
 Comparison with day 1 values: # = p<0.05, ## = p<0.01 and ### = p<0.001  
 Comparison between day 7 Vs day 30: \$ = p<0.05

Figure 1. Acrylic resins immersed in Turmeric and Yoghurt solutions showed a statistical significant color difference on day seven (p<0.01) and day 30 (p<0.001) when compared with base line and day one values. The color stability also differed (p<0.05) in these group between day seven and day 30. Acrylic resins immersed in tea solution also failed to retain its color on day 30 (for base line comparison p<0.001 and for day one comparison p<0.01) as their values statistically differed. However, this effect was not observed on day seven (p>0.05). There was also a difference in value when compared between day seven and day 30 (p<0.05). Acrylic resins immersed with coffee, cocoa and lime had similar effect. Their color stability was altered only on day 30 when compared with base line or day one values (p<0.05).

To summarize, the effect of staining solution was maximum in turmeric and yoghurt, followed by tea, coffee, cocoa and lime. It is also observed that turmeric, yoghurt solutions can alter the color stability even at seven days of immersion, however the remaining staining solutions exerted their effect only after 30 days of immersion but not after seven days of immersion.

**b) With HCS samples**



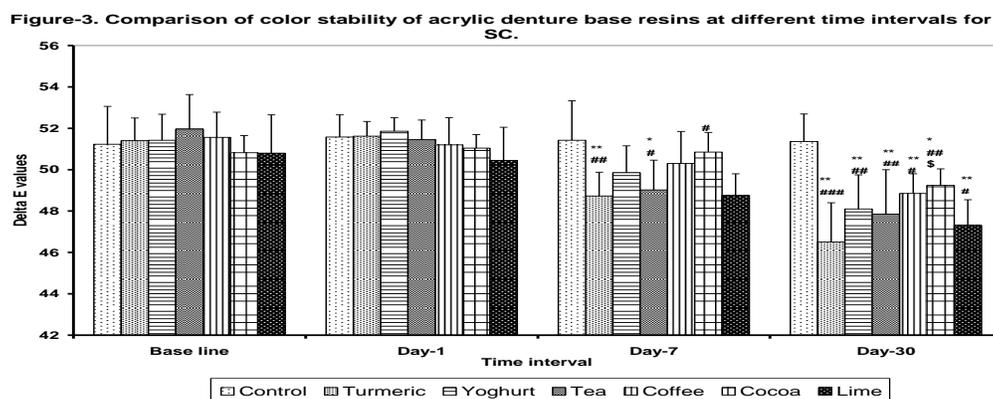
Comparison with base line values: \* = p<0.05, \*\* = p<0.01 and \*\*\* = p<0.001

Comparison with day 1 values: # = p<0.05, ## = p<0.01 and ### = p<0.001

Comparison between day 7 Vs day 30: \$ = p<0.05

Figure 2. Acrylic resins immersed in Turmeric (p<0.01), Yoghurt (p<0.05), Tea (p<0.01), Coffee (p<0.01) and Lime (p<0.05) solutions showed a statistically significant color difference on day 30 when their values are compared with either base line or day one values. It is also observed that acrylic resins immersed with coffee solution showed discoloration only after seven days of immersion (p<0.05). To summarize, the effect of staining solution was maximum in turmeric, coffee and tea followed by yoghurt and lime.

**c) With SC samples**



Comparison with base line values: \* = p<0.05, \*\* = p<0.01 and \*\*\* = p<0.001

Comparison with day 1 values: # = p<0.05, ## = p<0.01 and ### = p<0.001

Comparison between day 7 Vs day 30: \$ = p<0.05

Figure 3. Acrylic resins immersed in Turmeric solution showed a statistically significant color difference (p<0.01) on day seven as well as day 30 when compared with base line or day one values. Acrylic resins immersed with tea solution was also showed a statistically significant

color difference on day seven ( $p < 0.05$ ) and day 30 ( $p < 0.01$ ) when compared with base line or day one values. Yoghurt, Coffee and Lime had similar effect ( $p < 0.01$ ) on color stability on day 30 when their values are compared with either base line values or day one values. Immersion with Cocoa solution showed least significant ( $p < 0.05$ ) for 30 days of immersion when compared with either base line or day one values.

### **Comparison with different staining agents**

#### **a) With HCF samples**

The color stability of the acrylic resins was compared between different staining solutions at different time interval. The base line, day one and day seven values did not show any statistically significant difference ( $p > 0.05$ ). On immersion for 30 days, the color stability differed between control and turmeric ( $p < 0.001$ ), between control and tea; turmeric and cocoa ( $p < 0.01$ ), between turmeric and yoghurt, tea and cocoa ( $p < 0.05$ ). These data indicates that even after 30 days of immersion the color stability differs between different staining solutions.

#### **b) With HCS samples**

The base line, day one and day seven values did not show any statistically significant difference ( $p > 0.05$ ). On immersion for 30 days, the color stability differed between control and turmeric ( $p < 0.01$ ), control and tea ( $p < 0.01$ ), turmeric and cocoa ( $p < 0.05$ ) and tea and cocoa ( $p < 0.01$ ).

#### **c) With SC samples**

The base line, day one and day seven values did not show any statistically significant difference ( $p > 0.05$ ). On immersion for 30 days, the color stability differed between control and turmeric ( $p < 0.001$ ,  $F = 5.476$ ), control and yoghurt ( $p < 0.05$ ), control and tea ( $p < 0.05$ ) and control and lime ( $p < 0.01$ )

### **Comparison between different methods of curing**

Figure 4 and 5. Comparison of color stability of acrylic resins cured with different mode of curing did not show any significant change in color except for turmeric and yoghurt for seven days of immersion and coffee and cocoa for 30 days of immersion.

Acrylic resins cured with slow heat treatment (HCS) showed more color stability after immersion in turmeric solution for seven days period when compared with acrylic resins cured with fast heat treatment ( $p < 0.05$ ) and self cured resins ( $p < 0.05$ ,  $F = 6.083$ ). Similar result was observed for yoghurt solution for seven days of immersion. Acrylic resins immersed with yoghurt solution showed greater color stability ( $p < 0.05$ ) when compared with self cured resins immersed with yoghurt solution for seven days ( $F = 5.458$ ).

Figure 4: Comparison of color stability of acrylic denture base resins wrt different types of curing after 7 days of immersion period.

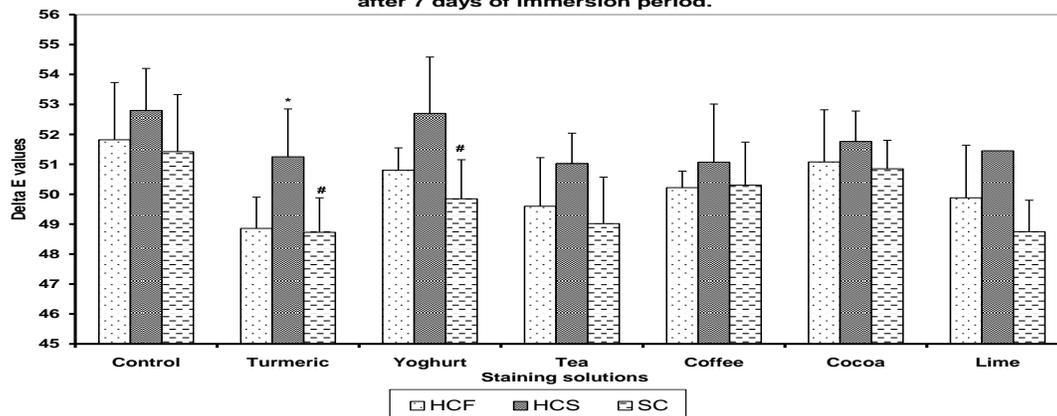
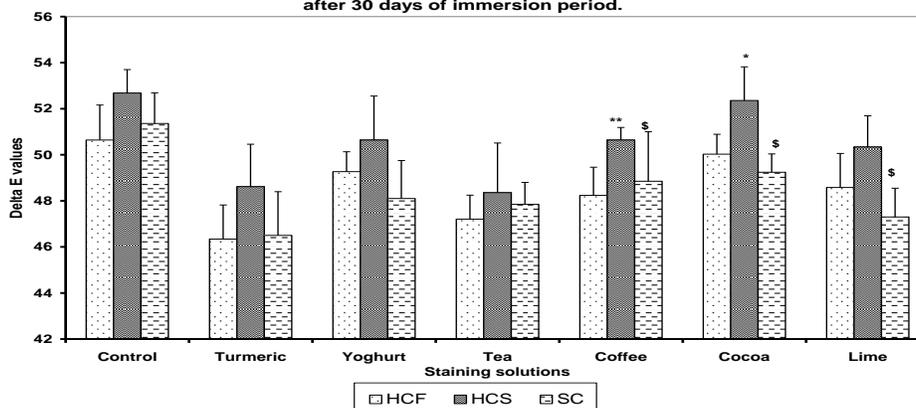


Figure 5: Comparison of color stability of acrylic denture base resins wrt different types of curing after 30 days of immersion period.



Comparison between HCF Vs HCS: \* =  $p < 0.05$ , \*\* =  $p < 0.01$  and \*\*\* =  $p < 0.001$   
 Comparison between HCF Vs SC: # =  $p < 0.05$ , ## =  $p < 0.01$  and ### =  $p < 0.001$   
 Comparison between HCS Vs SC: \$ =  $p < 0.05$ , \$\$ =  $p < 0.01$ , \$\$\$ =  $p < 0.001$

For 30 days of immersion with coffee, there was a statistically significant difference between acrylic resins cured with fast and slow heat ( $p < 0.01$ ) and also between heat cure slow resins with self cured resins ( $F = 11.241$ ). Immersion with cocoa solution also showed significant difference ( $p < 0.05$ ) between self cured resins and slow heat cured resins ( $F = 6.268$ ).

To summarize, it is clear from the data available that acrylic resins cured with slow heat treatment is more stable for color, especially against turmeric, yoghurt, coffee and tea solutions.

### DISCUSSION

The ultimate success of any prosthesis is when esthetic and functional excellence is achieved after meticulous attention at every stage of treatment.

In the present study, among all the staining agents used, Turmeric showed highest color changes in day 30 time interval ( $p < 0.001$ ). It is said that major constituents of turmeric

(Curcuma) are curcuminoids, the yellow coloring principles that cause stain. Smaller molecular size of curcumin coupled with the water absorption characteristics of the tested materials has created a stronger staining effect as discussed by Ergun et. al [4].

When considering the discoloration in coffee and tea, the specimens immersed in tea showed more discoloration. This is in agreement with the other studies in literature [16, 17]. Um and Ruyter et. al. [16] reported that the tea produced a yellow- brown stain while coffee stain was yellowish. The discoloration in tea was mainly due to surface adsorption of polar colorants at the surface. However other studies [3, 18, 19] found that coffee was more chromogenic than tea. The discoloration by coffee is due to both surface absorption and adsorption of colorants. Fine coffee particles deposits into pits of polymethylmethacrylate. The pits may have formed due to polymerization shrinkage of resin during curing. The less polar colorants and water soluble polyphenols in coffee for eg. Tannin, caffeine, caffeinic acid might have penetrated deep into the material, possibly because such colorants could be more compatible with polymer matrices.

When cocoa was considered there was less change in discoloration values. This result is attributed to the removal of accumulated layers. As the cocoa layers on specimen reached a certain thickness they tend to break away from the surface of samples and return to the solutions. This was the reason for less staining characteristics in cocoa.

Polymeric materials reportedly display a tendency to erode under acidic conditions [10, 20]. In general, food stuffs with low pH have greater erosive effect. Low pH affected the surface integrity of polymers. This was because under acidic conditions, the polymer surface was appreciably softened by loss of structural ions. Yoghurt and lime exhibited a similar behavior. The acidic ingredients in yoghurt and lime for eg. lactic acid and citric acid have caused surface dissolution of polymeric surface leading to much paler appearance. Hence specimens after their specified immersion period when observed visually showed a lighter color match when compared to control.

Amongst the various curing procedures that were followed specimens cured by HCS method was most color stable when compared to HCF, whereas SC samples showed least color stability. Literature study [21-23] reveals that the color stability of chemically activated denture acrylic resins have been found to be less stable than conventional acrylic resins. This color change was associated with the chemical composition of the monomer, type and quantity of amine involved in polymerization and inhibitor used. This study was thus parallel to the findings with the studies found in literature. Austin et. Al [15] explained that denture base materials processed by a cold polymerized method have demonstrated upto seven times the level of residual monomer found in conventional heat polymerized materials. The residual monomer content was responsible for the color changes observed. Acc to May et.al.[24] color changes may be associated with porosity caused by overheating or insufficient pressure during polymerization. Self cure demonstrated highest color difference for the material category. Self cure resins were fabricated with minimal stirring of mixing and in same fashion as the other polymethyl methacrylate polymer and monomer, yet it demonstrated air inclusions which were

evident after polishing. The inclusions may provide reservoirs for moisture and stain to settle and could contribute to color changes noted in this study.

Extrapolating from the results of this *in vitro* study it can be said that the present study was inline with the hypothesis to be tested before the study. This reveals that color stability in acrylic denture base resins was significantly influenced by type of materials, method of cure, immersion periods, staining agents and dietary habits of patients. However, to investigate the color stability performance of DBR in a clinical setting, these results should be supported by planned *in vivo* studies.

### CONCLUSION

From the present study we can conclude that, color stability is of great importance to patients and clinicians when working in the esthetic zone. Patients should be aware of their dietary habits if their prosthesis needs to be worn for long period and therefore, may be advised to avoid or minimize consumption of these beverages during the service of the DBR. The dentist should select and use materials with good color stability, for the excellent serviceability of the prosthesis.

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