Color stability of chemically activated reline resin after microwave disinfection: A 1-year clinical trial

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ABSTRACT: Purpose: To evaluate the effect of microwave disinfection on the color stability of a hard chairside reline resin after a 1-year service period. Methods: 40 adult patients aged between 30-75 years, who required denture reline treatment, participated in this study. Tokuyama Rebase II was used to reline complete maxillary dentures. The edentulous subjects were randomly divided into two groups (n=20) and dentures were cleansed according to two methods: CG (control group) - brushing with coconut soap and soft toothbrush; DG (disinfection group) - brushing according to previous methods and microwave disinfection once a week for a week at 650W. Color parameters in L*a*b* were recorded by spectrophotometer immediately after the reline, at 7 and 15 days, 1, 3, 6 and 9 months and 1 year post-placement. Data were analyzed by two-way repeated-measures ANOVA and Tukey tests (α< 0.05). Results: Color alteration values of DG were significantly lower than those of CG (P< 0.05). Color changes observed after 15 days were greater than values obtained at 7 days recall (P< 0.05). All color changes observed for the CG were considered noticeable (between 1.5 and 3.0 NBS). In DG, color change was slight (between 0.5 and 1.5 NBS). There were statistically significant differences between L* values obtained initially and after 3 months, between 15 days and 3 months and between 15 days and 1 year (P< 0.05). No significant differences were observed between group and time for the parameters a* and b*. (Am J Dent 2011;24:200-204).

CLINICAL SIGNIFICANCE: Microwave disinfection caused improvement in color stability of the reline material.

Introduction

It is known that hard direct relining is used to improve retention, support and stability of removable prostheses. This procedure can reproduce the morphologic features of oral soft tissue directly on the denture base, improving loss of denture fit that is the patient’s chief complaint over time. Therefore, periodic assessment and reestablishment of fit by relining procedures can extend the long-term performance of dental prostheses. Nonetheless, when relined dentures are subjected to prolonged exposure to staining solutions in the oral environment, resin color is affected by several discoloring agents arising from a number of extrinsic and intrinsic factors. The color of resin-based materials may change over time because they are prone to absorption and adsorption of liquids. Other factors also have an effect on the degree of color stability, such as incomplete polymerization, water sorption, chemical reactivity, diet, oral hygiene, surface roughness, stain accumulation, dissolution of ingredients, degradation of intrinsic pigments, and the chemical composition of the monomer. Despite the denture reline materials used on surfaces that are not clinically visible, their color should be esthetically acceptable and similar to that of the denture base material. Material that undergoes detectable color changes may be a source of embarrassment for both the patient and dentist, requiring dentures to be replaced, with further costs.

One study described the color stability of denture base acrylic resins and hard direct reline acrylic resins when exposed to food colorants. McNeme et al reported that the use of a denture cleanser (sodium hypochlorite) caused bleaching of acrylic resin. May et al noted that the color stability of seven conventional and one microwave heat-polymerized denture base materials was affected after accelerated aging. Hong et al showed that the color stability of denture base acrylic resins was influenced by the type of polymerization and the type of denture cleanser used. However, none of these reports studied the influence of microwave disinfection on this process.

Studies have shown that microwave irradiation is an effective method to kill microbial biofilms formed on acrylic resin dentures. Although other laboratory studies evaluated the effects of microwave disinfection on the hardness, bond strength to denture base resins, flexural strength, porosity, and dimensional stability of denture base resins, there is a lack of long-term clinical studies assessing the color stability of reline materials after microwave disinfection.

Considering that one of the factors that determines the clinical longevity of a material is its ability to maintain its original color when functioning, this randomized controlled clinical study evaluated the effect of microwave disinfection on the color stability of reline dentures during a service period of 1 year.

Materials and Methods

This prospective randomized study was conducted between March 2006 and July 2009. The study population consisted of 40 subjects who were attending the School of Dentistry of Araquara, UNESP. The treatment protocol (31/05) was approved by the Ethics Committee of the Araquara Dental School, São Paulo State University and each patient provided written consent to participate in this study. The subjects were healthy and their maxillary dentures required relining. Tokuyama Rebase II was used to reline complete maxillary dentures. From each denture, material from the tissue side...
little over the border was removed with a tungsten-carbide rotary cutting instrument to provide space for an adequate thickness of reline material. The intaglio surface of the denture base was cleaned and dried. A coat of a separating agent was applied over the polished surfaces and artificial teeth. The bonding agent provided by the reline resin manufacturer was applied with a brush on the intaglio surface of the denture base, and the reaction time (120 seconds) occurred before the resin was inserted. Tokuyama Rebase II was proportioned and manipulated following the manufacturer’s instructions. The material was applied in a uniform layer on the tissue side of the denture base and over the borders, and the denture was immediately placed in the patient’s mouth in its terminal position. Afterwards, with the patient’s mouth open, the cheeks were manipulated to turn the excess at the border and establish harmony with bordering attachments. Before initial hardening, the denture was removed from the mouth and gross excess material was removed with scissors. After this the denture was replaced in the patient’s mouth in its terminal position, and the teeth were brought into occlusion until setting was complete. After final hardening, the relined dentures were adjusted.

After relining, the baseline color of all dentures was determined in the spectrophotometer Color Guide 45/0° according to the CIE (Commission Internationale de l’Eclairage) L*a*b* system. Color measurements were made on the convex area of the intaglio surface of removable dentures through the sighting device for samples provided by the manufacturer of the spectrophotometer that has a circular hole 15 mm in diameter. The standardization of the color measurements was obtained through the coincidence of a line made on that sighting device with the labial notch region (Figure). All evaluations were conducted by two examiners working separately and means for the L*, a* and b* values were calculated. After baseline color measurements were made, the patients were randomly divided into two groups (n= 20) and allocation concealment was by the use of sealed, numbered envelopes. The assignment was balanced with the use of blocks of four. The dentures were cleansed according to one of two methods: CG (control group) - brushing with coconut soap and soft toothbrush; DG (disinfection group) - brushing according to previous methods and microwave disinfection once a week during the entire evaluation period (1 year). The dentures were subjected to disinfection in a domestic microwave oven calibrated to 650W, for 3 minutes, with the specimens immersed in 200 ml of water during the irradiation time.

Each relined denture was subjected to evaluation after 7 and 15 days, 1, 3, 6 and 9 months and 1 year. The total color change (ΔE*) of each reline denture was then calculated using the relationship:

$$\Delta E^* = (\Delta L^* + \Delta a^* + \Delta b^*)^{1/2}$$

All study team members were blinded until study completion, except the clinician who performed the relining and disinfection. Furthermore, in the last evaluation, all volunteers answered a questionnaire on daily habits to investigate the influence of the patient’s habits on the staining of the reline resin. The questionnaire contained questions as follows:
- Are you a smoker? How many times per day?
- Do you drink coffee? How many times per day?
- Do you drink tea? How many times per week?
- Do you drink cola-based soft drinks? How many times per week?

The statistical analysis was designed to study any color change in reline material after microwave disinfection. Comparison between L*, a* and b* baseline values for both groups was undertaken using the non-parametric Wilcoxon test. A transformation of the data was applied to achieve a more normal distribution for each variable, which is desirable when using parametric statistical procedures. Ln transformation was performed on ΔE*, L*, a* and b* values and a potency transformation (b25) was applied for b* data. Levene and Shapiro-Wilk tests (Statistica 6.0®), were applied to test homogeneity of variance and normality, respectively. The results of ΔE*, L*, a* and b* were evaluated statistically by two-way repeated-measures ANOVA. The two factors analyzed were group and time after relining. Tukey Honestly Significant Difference (HSD) post hoc test was used to determine differences between means (α = 0.05).

Descriptive statistics were used to report the daily habits of the volunteers who answered the questions.

### Results

Descriptive statistics analysis (Table 1) and Wilcoxon’s test were used to compare the L*, a* and b* baseline values between
control and disinfection groups. Immediately after relining, no significant difference was found between baseline values of the two groups. This finding indicates that the groups were statistically similar prior to treatment.

Comparisons between groups and time after relining were made by two-way repeated-measures ANOVA after natural logarithmic (ln) transformation of ΔE*, L* and a* values and potency transformation (b*0.20) for b* data, where P< 0.05 was considered significant. Further post tests were done by Tukey’s analysis.

According to the ANOVA results for the ln(ΔE*) values, it can be seen that significant differences were found for the 2 main factors, “group” and “time after relining” (P< 0.0001). The two-way interaction was not significant. The two-way repeated-measures ANOVA for ln(L*) values demonstrated significant differences for “time after relining” (P< 0.0003). ANOVA for ln(a*) and (b*0.20) showed that there were no significant differences between the “group” and “time after relining” factors and the interaction.

Table 2 presents the mean values for ln(ΔE*) and the results of Tukey HSD post hoc test (α= 0.05). Microwave disinfection produced specimens with significantly lower color change values irrespective of the time, when compared with specimens that were not irradiated (P< 0.0001). The two-way ANOVA and Tukey HSD post hoc tests showed significant differences among the “time after relining” (P< 0.0001) irrespective of evaluated group. Color change values observed at 1 month, 3, 6 and 9 months and 1 year were higher than the values obtained at 7-day recall (P values of 0.008, 0.005, < 0.0001, 0.005 and < 0.0001, respectively).

Table 2 also presents the mean values for ln(L*) values and the results of Tukey HSD post hoc test (α= 0.05). There were statistically significant differences between L* values obtained initially and during the course of 3 months (P< 0.05), between 15 days and 3 months (P<0.0281) and between 15 days and 1 year (P= 0.0324).

The National Bureau of Standards (NBS) established a rating system to describe color differences in NBS units. NBS units are expressed by the following formula:

NBS unit= ΔE* x 0.92

All color changes (Table 3) observed for the CG were considered noticeable (between 1.5 and 3.0 NBS). In disinfection group, color change was slight (between 0.5 and 1.5 NBS).

With respect to the daily habits, 15% of the volunteers said they smoked 5.66 times per day. Twenty percent of volunteers drank tea once a week and 24 patients (55%) drank cola-based soft drinks weekly, and of these, 16 patients ingested the drink twice a week.

**Discussion**

Color change can be evaluated by visual and instrument determination. However, spectrophotometers and colorimeters have provided consistent color evaluation, because instrumental readings eliminate the subjective interpretation of color comparison. The color of dental materials is often expressed in L*, a*, b* coordinates (CIE Lab Color system), a method developed in 1978 by the Commission Internationale de l’Eclairage for characterizing color based on human perception. This system represents three-dimensional color space and consists of coordinates L*, a* and b*. The L* represents the lightness and is located along a vertical axis that ranges from a value of 0 for black to 100 for perfect white. The color coordinates a* and b* represent a position between red (+a*) and green (-a*), and between yellow (+b*) and blue (-b*). The a* and b* coordinates revolve on axes around L*. Spectrometric color measurement generally uses the illuminant D65 to simulate daylight. The differences in the lightness and chromaticity coordinates are calculated (ΔL*, Δa*, Δb*) and the total color change will be represented by distance between the initial position (the initial match of L*a*b* coordinates) and the spatial position obtained in each measurement.

Color change in the oral environment may occur due to variability of intrinsic and extrinsic factors. The initiator, degree of conversion and water sorption are intrinsic factors that play an essential role in the color stability of the resin-based materials. Extrinsic factors include adsorption and absorption of pigments...
derived from dental biofilm, smoking and dietary habits. 

The results of this study showed that dentures relined with Tokuyama Rebase II and submitted to microwave disinfection presented lower color changes than the control group (CG). Auto-polymerizing relining acrylic resin has a higher residual monomer content when compared with heat-polymerized acrylic resins used for denture base materials. Therefore, high levels of residual monomer are likely to remain after polymerization. The rise in temperature during microwave disinfection may have promoted further monomer to polymer conversion. Furthermore, when the dentures were microwaved, the rise in temperature may have increased the diffusion rate of this unreacted monomer into water and its vaporization. With regard to the efficiency of polymerization, it is important to note that the higher the degree of conversion, the less the residual monomer available to produce color-degraded products. One can suppose that the lower content of residual monomer obtained after microwave irradiation resulted in lower rates of oxidation, and consequently, in a decrease in the intrinsic color changes.

Color change was observed after 1 month, 3 months, 6 months, 9 months and 1 year when compared with 7 days. These results are in agreement with Hong et al., who observed an increase in color change of acrylic resins over a long period of time. The susceptibility to color change of the material evaluated in this study can be attributed to the degree of water absorption and hydrophilic characteristic of the resin. The monomers used in the Tokuyama are 2-acetoacetoxyethyl methacrylate and 1,9-nonanediol dimethacrylate and these constitute the main liquid components of the material. The relining material powder consists of polyethyl methacrylate. The hydrophilicity of polyethyl methacrylate may elucidate the staining observed over time. Moreover, water sorption can explain the uptake of fluids, resulting in lower values for hydrophobic materials. It was also reported that hydrolytic degradation through the formation of pores on the surface of the resins may allow stain penetration and discoloration. Acrylic resin porosity is a common clinical occurrence, which could result from vaporization of the unreacted monomer, air trapped during mixing and monomer shrinkage during polymerization. Porosities inside relining material may also have facilitated the sorption of liquids. Furthermore, another predisposing factor for staining of relined denture is roughening of the material caused by copying the palatal tissue surface. In this study, a period of 1 month when compared with 7 days was sufficient to induce color change in the relined denture.

It is important to emphasize that irreversible endogenous discoloration may occur as a result of the aging of relining material and changes in the structure of the material, or internal discoloration due to the incomplete conversion of initiators and unconverted C = C bonds. Another hypothesis is that the extrinsic discoloration of the evaluated resin may have occurred due to surface absorption of food dyes or mouthwash, smoking or the presence of biofilm.

Patients’ dietary habits may have a significant influence on the color of dental materials. A high frequency intake of coffee, tea and soft drinks may increase pigmentation of teeth and dentures. In this study, the answers to the questions indicated that the patients often drank coffee, tea or cola. The coloring found in these beverages (coffee, cola and tea) may have caused an increase in the values of the L* after a long period of time. Coffee has brown pigments, derived from caramelization and Maillard reactions (chemical interactions between an aldehyde or ketone group of a reducing sugar and amino group, resulting in brown polymers called melanoids), which occur during the coffee bean roasting process. The products of caramelization and Maillard reactions are the caramel and melanoids respectively, responsible for the color of coffee and chromogenic effect of this beverage on dental materials. The dark brown color observed in cola is also due to caramels. Tea acquires its brown color during processing due to the conversion of chlorophyll into phaeophorbides (brown) and phaeophriths (black). Therefore, the dentures probably became browner.

The subjects in this research were instructed not to use denture cleansers or mouthwash. Thus, relining material did not show significant alterations for color parameters a* and b* that could be caused by green, blue or red pigments added to these solutions to make them more attractive to consumers.

With reference to smoking, 15% of the volunteers were smokers. Among the components of cigarettes are nicotine and tar. According to Takeuchi, tar is responsible for resin composite material staining, since pure nicotine is an alkaloid contained in the tobacco leaf, which in liquid form is colorless. Whereas the color of tar ranges from brown to black, Belli et al evaluated the effect of cigarette stains on materials used in veneers. They concluded that cigarette smoke had the greatest potential for staining followed by coffee, tea and distilled water. There was visible color change probably related to cigarette smoking.

Color change of extrinsic origin may also be due to the presence of biofilm on the denture relining. The deposition of salivary proteins and food on denture surface with the consequent formation of biofilm makes color change possible by facilitating the adhesion and absorption of staining. The limitations of this clinical study include the incapacity to fully control the patient’s dietary habits and thus assess the real potential of staining foodstuffs. Furthermore, the effect of denture cleansers on the color stability of the relined dentures should be evaluated in future clinical studies.

Based on all these considerations, the microwave irradiation can be used for disinfection of relined dentures without causing deleterious effects on the color of the evaluated material. Moreover, a long period of denture wearing may cause discoloration of the materials due to the combination of intrinsic and extrinsic effects.
References