# *Effect of Aging and Mouthrinses on the Color Stability of All-Ceramics*

# *Yaşlanma ve Ağız Gargaralarının Tam Seramiklerin Renk Stabilitesi Üzerine Etkisi*

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***ÖZET***

***Amaç:*** *Mevcut in vitro çalışmanın amacı, tam seramik malzemelerin renk stabilitesi üzerinde ağız gargaraları ve termal döngünün etkisini değerlendirmektir.*

***Gereç ve Yöntem:*** *Toplamda 100 adet 1,5 mm kalınlığında tam seramik numune 0,5x10x10 mm'lik alt yapılara dentin porseleni uygulanarak hazırlandı. Renk verileri spektrofotometre ile kaydedildi ve renk farklılıkları CIEDE2000 formülü ile hesaplandı. Başlangıç ölçümlerinden (T0) sonra örnekler 2 eşit gruba ayrıldı (Grup A ve Grup B). Grup A'ya ait numuneler termal döngü cihazında yaşlanmaya maruz bırakıldı ve renk değişimleri kaydedildi (T1). Her iki grup daha sonra 5 alt gruba ayrılarak (n:10) örnekler Klorhex® (KLO), Listerine Total Care® (LIS), Tantum Verde® (TAN), Meridol® (MER) ve distile su (DIS) içinde 12 saat bekletildi. Son renk değişimleri kaydedilerek (T2) renk farkı değerleri ($\Delta E_{00}$) istatistiksel olarak Kruskal-Wallis ve Mann-Whitney U testleri ile karşılaştırıldı.*

***Bulgular:*** *MER'in renk farklılıkları, LIS dışında grup A ve grup B'deki diğer gargaralardan istatistiksel olarak farklı bulundu ($p<0,001$). LIS her iki grupta da DIS'den istatistiksel olarak anlamlı farklılık gösterdi ($p<0.05$). Grup A'da MER'deki ortalama değerler algılanabilir sınırlar içinde olup, LIS'deki ortalama değerler kabul edilebilir sınırlar içinde belirlendi (T0-T2). Grup B'de gargaraların ortalama değerleri algılanabilir sınırların (T0-T2) altında hesaplandı. Termal döngü ile yaşlandırma istatistiksel olarak anlamlı bir renk farklılığına (T0-T1) neden olmadı.*

***Sonuç:*** *Ağız gargaralarının uzun süreli kullanımı tam seramik malzemelerde renk değişikliğine neden olabilir. Amin/kalay florür içeren ağız garagaraları (MER) günlük kullanım için dikkatle önerilmelidir.*

***Anahtar Kelimeler:*** *Seramik; renk stabilitesi, ağız gargarası; termosiklus, yaşlanma*

***ABSTRACT***

***Aim:*** *The aim of this in vitro study was to evaluate the effect of mouthrinses and thermocycling on the color stability of all-ceramic materials.*

***Materials and methods:*** *A total of 100 all-ceramic samples with a thickness of 1.5 mm were prepared by applying dentin porcelain to the substructures within 0.5x10x10 mm dimensions. Color data were recorded with a spectrophotometer and color differences were calculated with CIEDE2000 formula. After baseline measurements (T0), the samples were equally divided into 2 groups (Group A and Group B). Samples in group A exposed to aging in a thermocycling device and color changes were recorded (T1). Groups were then divided into 5 subgroups (n:10) and the samples were immersed in Klorhex® (KLO), Listerine Total Care® (LIS), Tantum Verde® (TAN), Meridol® (MER) and distilled water (DIS) for 12 hours. The last color changes were recorded (T2) and the color difference values ($\Delta E_{00}$) were statistically compared with the Kruskal-Wallis and Mann-Whitney U tests.*

***Results:*** *MER was statistically different from other mouthrinses except for LIS ($p<0.001$). LIS was statistically different from distilled water in both groups ($p<0.05$). In group A, the mean values in MER were within the perceptible limits and LIS were within the acceptable limits (T0-T2). The mean values of mouthrinses in group B were below the perceptible limits (T0-T2). Thermocycling did not cause a statistically significant color difference (T0-T1).*

***Conclusion:*** *Prolonged use of mouthrinses may cause color change in all-ceramic materials. Mouthrinses containing amine/stannous fluoride (Meridol®) should be recommended with caution for daily use.*

***Keywords:*** *Ceramics, Color stability, Mouthrinse, Thermocycling, Aging*

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**INTRODUCTION**

There is now a growing interest and rapid development about ceramic restorations that do not contain metal substructures, according to the aesthetic restoration expectations among patients. Traditional ceramics are biocompatible and aesthetic materials, but they have a fragile structure with low tensile strength. These disadvantages of traditional ceramics allow the development of materials with high mechanical and biological properties, such as zirconia crowns.[1,2]

Biocompatible aesthetic results and high mechanical properties are obtained by using zirconia as a substructure material in full-crown restorations.[3] Zirconia is a polymorphic material, so it can exhibit multiple crystal structures depending on temperature and pressure conditions. There are 3 forms of zirconia; monoclinic, tetragonal, and cubic phase.[4,5] 3 mol% yttrium oxide ($Y_2O_3$) is added to stabilize the zirconia at room temperature in the tetragonal phase. In this way, yttrium tetragonal zirconia polycrystalline (Y-TZP) material with superior mechanical properties is obtained.[6,7] Despite its mechanical properties, the opaque structure of Y-TZP is a significant disadvantage. For this reason, porcelain is layered on the zirconia substructure to achieve aesthetic results.[7]

The color changes that occur during restorations are a problem for both patients and dentists. The color of restorative materials can be affected by plaque accumulation, coloring solutions, surface roughness, and chemical degradation. The consumption of various foods and beverages can cause physico-chemical reactions in the matrix in the superficial and deep layers of restorative materials, which, in turn, can cause changes in the structural properties of the materials.[8-11]

The efficient removal of dental plaque is essential for maintaining oral health, as plaque has long been identified as a critical factor in the etiology of caries, gingival inflammation, and chronic periodontitis.[12,13] Toothbrushing is generally accepted as the most efficient oral hygiene method of cleaning one's teeth. Patients' efforts, however, are often compromised by the presence of hard-to-reach areas as well as inadequate skill, poor motivation and lack of compliance. Consequently, the use of antimicrobial mouthrinses has been proposed as adjuncts to mechanical oral hygiene regimens and is considered a mean to enhance plaque removal.[14,15] Despite the antimicrobial effect of mouthrinses, frequent usage of these may have unwanted effects on the surface properties of restorations resulting in discoloration in restorative materials.[16,17]

Mouthrinses contain various component such as antimicrobial agents, salts, organic acids, dyes, and in some cases alcohol.[18] It is known that the hydroxyl groups of alcohols react with the $Zr^{+4}$, $Si^{+4}$, and $Zn^{+2}$ cations of restorative materials to cause degradation of the materials. The F anion can interact with Si, the main element of bioceramic materials, and the $SiF_4$ component may be responsible for the corrosion of bioceramics.[19,20]

The mouthrinses in the present study were selected as being the most preferred ones in daily plaque control and as follows:

Chlorhexidine digluconate (CHX) is an antibacterial agent that is widely used as a mouthrinse. Following the use of CHX, side effects such as discoloration of tooth enamel or restorative materials, the formation of dental calculus, and unpleasant after taste have been reported. It is available on the market as a mouthrinse under the name Klorhex® (KLO).[21, 22]

Listerine® (LIS) is a mouthrinse used as an anti-plaque agent to treat gingivitis. Listerine consists of a suspension of essential oils, eucalyptol, thymol, menthol, and methyl salicylate in 26.9% alcohol.[23]

The mouthrinse available on the market under the name Tantum Verde® (TAN) contains

Benzydamine HCl. Benzydamine HCl is a non-steroidal anti-inflammatory drug (NSAID) that has systemic and locally applicable analgesic, antipyretic, local anesthetic effects.[24]

Meridol®, (GABA INT (alcohol-free version of Meridol®– 250 p.p.m. SnF2/AmF) (MER) have active ingredients of amin/stannous fluoride and zinc lactate to prevent tooth decay and inflammation of the gums as well as reduce plaque build-up.[25,26]

Thermocycling is one of the aging methods that was developed to mimic the physical effects caused by long-term clinical use of restorative materials in short periods and under stable conditions. The temperature values of thermocycling solutions were determined as 5°-55°C according to ISO standards and 10,000 cycles of aging with thermocycling was reproted to be equivalent to 1 year of in vivo use.[27]

Maintaining the color stability of restorations is important for ensuring aesthetic success. Therefore, the daily recommended mouthrinses should not affect the aesthetic properties of the restorations. There exists no available study that investigated the effects of MER on the color stability of all-ceramic materials. The present study aimed to evaluate the effects of four commonly used mouthrinses with different ingredients on the color stability of all-ceramic materials. The null hypothesis of our study was established on; the color changes caused by mouthrinses in all-ceramic materials will be significant, and the color changes caused by the thermocycling process in all-ceramic materials will be significant.

## MATERIALS AND METHODS

Four different mouthrinses in different formulations (KLO, LIS, TAN, MER) and standardized all-ceramics were used to evaluate the staining (Table 1).

**Table 1.** Mouthrinses used in our study.

| Mouthrinses | Composition | Manufacturer |
|---|---|---|
| Klorhex® | 0.2% Chlorhexidine Gluconate, Glycerin, Lemon Extract, Peppermint Extract. | Drogsan Pharmaceutical Ind. And Trade Inc., Ankara, Turkey |
| Listerine Total Care® | Alcohol, Benzoic Acid, Sorbitol, Eucalyptol, Sodium Flouride, Sodium Benzoate, Thymol, Menthol, Methyl Salicylate | Johnson and Johnson Sanitary Material Industry Trade Ltd. Comp., Istanbul, Turkey. |
| Tantum Verde® | 0.15% Benzydamine Hydrochloride, Methyl Paraben, Quinoline Yellow, Patent Blue V, Ethanol, Mint Flavor. | Angelini Pharmaceutical Ind. And Trade Inc., İstanbul, Turkey. |
| Meridol® | Amine Flouride, Stannous Fluoride, Aqua, Xylitol, Hydrogenated Castor Oil, Olaflur, Aroma, Sodium Saccharin, CI. | Colgate-Palmolive, Hamburg, Germany. |

The distilled water (DIS) was the rinsing solution for the control group. The sample size was calculated using G * Power V3.1.9.2 program. The effect size was calculated as 1.53 based on a similar study that compared the color change values of distilled water and listerine in all-ceramic materials.[28] According to this effect size, it was planned to recruit 10 samples to each subgroup (N=100) with a 95% confidence level and 95% study power. The flow chart of the study is summarized in Figure 1.

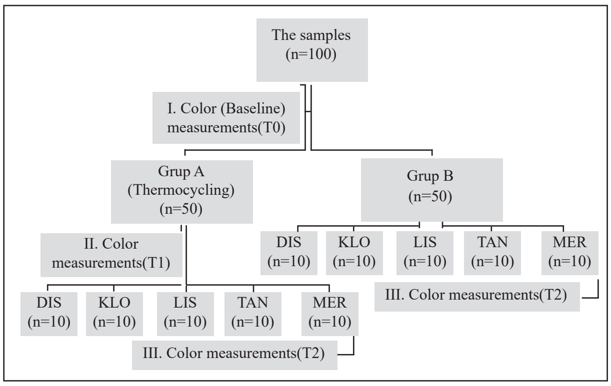


**Figure 1.** Flow chart of the study. DIS, distilled water(control); KLO, Klorhex; LIS, Listerine Total Care; TAN, Tantum Verde; MER, Meridol.

**Preparation of samples**

Pre-sintered Sirona-cerec inCoris ZI F1 monoS blocks (Sirona Dental Systems, Bensheim, Germany, LOT2015497929) with yttrium-stabilized zirconium oxide content were used as substructure materials. The substructures of the samples were prepared with an Isomet precision cutting device (Low-Speed Saw, Buehler Lake Bluff, IL USA) using a special diamond disc (Diamond Wafering Blade Series 15 HC Diamond No. 11-4244, IL USA) at low speed (300 m/s) under water cooling by the same investigator (BİK). The substructures of the samples were prepared in dimensions of 0,7x12x12 mm. All the substructures were sintered in accordance with the manufacturer's instructions for 92 minutes at 1581°C in a high-heat sintering furnace (Infire HTC Speed; Sirona, Germany). After sintering contraction, substructure samples in 0,5x10x10 mm were obtained. The thickness of the samples was measured with a digital micrometer (Astor, Digital Caliper, China). 600, 800, 1000, and 1200-grid SiC sandpaper (Buehler-Met II Silicon carbide grinding paper P400/600, USA) was applied underwater cooling to ensure surface smoothness and standardization of the substructures of the samples. To remove the residues remaining on the surfaces, the samples were cleaned for 10 minutes using distilled water and air-dried.

Kuraray Noritake Cerabien ZR dentin porcelain (Kuraray Noritake Inc., Japan) of A2 color was applied to all substructure samples by the traditional layering method. After the powder and liquid of dentin porcelain were mixed by the instructions on a cement glass, the resulting porcelain clay was applied to the substructures with the help of a brush. Then the samples were baked in the Programat P310 (Ivoclar, Vivadent, Liechtenstein) porcelain oven in accordance with the manufacturer's instructions.

After the porcelain was layered, correction operations were performed so that the total thickness of the samples was resulted to 1,5 mm. The surfaces of all samples were sanded again underwater cooling. All samples were cleaned for 10 minutes using distilled water in an ultrasonic cleaner and air-dried. Then, a thin layer of glaze (Kuraray Noritake CZR FL, Japan) material was applied to the upper surfaces of the samples. Prepared samples were randomly selected and divided into two groups (Group A and Group B). Each group was divided into 5 subgroups and the samples were given numbers from 1 to 10.

**Experimental Procedure and Calculation of $\Delta E_{00}$ values**

All samples were immersed in distilled water for 24 hours before the first color measurements. After that, baseline color measurements of the samples (T0) were performed with the SpectroShade (MHT Optical Research, Niederhasli, Switzerland) device in the Laboratory of the Faculty of Dentistry of Ondokuz Mayıs University. The L*a*b* color data of the samples were recorded. In order to ensure standardization in the measurements, the silicon index was used and measurements were made from the glazed surfaces of the samples. The spectrophotometer device took measurements from three different points for each sample and presented the average value of these measurements. Calibration of the device was achieved by taking measurements of the white and green colors on the device box in accordance with the manufacturer's instructions.

Followed by the baseline color measurements, fifty samples were aged with 10,000 cycles in a thermocycling device (Thermocycler SD Mechatronics, Feldkirchen-Westerham· Germany). After the thermocycling, the second color (T1) measurements of 50 samples were performed with a spectrophotometer. The L*a*b* color data of the samples were recorded. In our study, samples in groups A and B were immersed in 5 different mouthrinses for 12 hours to coincide with 1 year of mouthwash use.[29] Then all samples were washed with distilled water and air-dried. Distilled water was used as a control group in our study. The color measurements (T2) after immersion of all samples in mouthrinses were performed with a spectrophotometer. The L*a*b* color data of the samples were recorded.

The CIEDE2000 formula was previously reported to better fit for the calculated color differences and reflects the color difference better than the CIEL*a*b* formula.[30] Therefore, The CIELab values were converted into CIEDE2000 L', C' (chroma), h' (hue) values, while $\Delta E_{00}$ color differences were calculated using the following formula:

$$\Delta E_{00}=\sqrt{\left(\frac{\Delta L'}{K_L S_L}\right)^2+\left(\frac{\Delta C'}{K_C S_C}\right)^2+\left(\frac{\Delta H'}{K_H S_H}\right)^2+R_T\left(\frac{\Delta C'}{K_C S_C}\right)\left(\frac{\Delta H'}{K_H S_H}\right)}$$

where ΔL', ΔC', ΔH' indicates the differences between the corresponding color coordinates computed based on the uniform color space used in CIEDE2000, and KLSL, KCSC and KHSH are empirical terms for converting the differences for each coordinate into the CIEDE2000 difference formula.[31] In the present study, the parametric factors of the CIEDE2000 color-difference formula were taken as 1.[32] The value for perceptibility was set as $\Delta E_{00} \leq 1.30$ and for clinical acceptability threshold as $\Delta E_{00} > 2.25$.[33]

**Statistical Analysis**

The data obtained were analyzed statistically using the SPSS 22.0 (SPSS version 22.0 software, SPSS Inc., Chicago, IL, USA) Windows program. The values of $\Delta E_{00}$ were compared with the non-parametric Kruskal-Wallis and Mann-Whitney U tests. The group that made the difference was determined by the Dunn test. The L*, a*, and b* values were compared with the analysis of Variance in repeated measurements as they showed a normal distribution. Mann-Whitney U test was used to compare $\Delta E_{00}$ values in terms of thermocycling groups (Group A-B). The significance level was taken as $p<0.05$ in all tests.

## RESULTS

In group A, MER was found to be statistically different from DIS and TAN in T0-T2 measurements ($p<0.001$). LIS was also different from DIS ($p<0.05$). The mean $\Delta E_{00}$ values of the samples in the MER were found within the perceptible limits, and the mean values in the LIS were within the acceptable limits (Table 2).

There was no statistical difference within the $\Delta E_{00}$ values of the samples of T0-T1 measurements in group A (p=0,097). Thermocycling did not cause a statistically significant color change in the samples (Table 2).

In Group B, MER was statistically different from DIS, KLO and TAN ($p<0.001$). LIS was found to be different from DIS ($p<0.05$). When the mean $\Delta E_{00}$ values of the groups were examined, it was observed that the color change of the samples in all mouthrinses was below the perceptible limits (Table 2).

**Table 2.** Comparison of $\Delta E_{00}$ (Mean values±Standard deviation)

| | DIS | KLO | LIS | TAN | MER |
|---|---|---|---|---|---|
| **Group A (T0-T2)** | 0,40±0,14[a] | 0,55±0,23[abc] | 1,82±2,82[bcd] | 0,50±0,24[abd] | 1,30±0,66[c] |
| **Group A (T0-T1)** | 0,32±0,10 | 0,51±0,20 | 0,54±0,22 | 0,45±0,25 | 0,37±0,28 |
| **Group B (T0-T2)** | 0,34±0,15[a] | 0,43±0,15[ab] | 0,96±0,66[bc] | 0,47±0,37[ab] | 1,15±0,27[c] |

DIS, distilled water (control); KLO, Klorhex; LIS, Listerine Total Care; TAN, Tantum Verde; MER, Meridol; a, b, c, d indicate statistical differences among the mouthrinses; *p<0,05.

There was no statistical significance between the color changes caused by each mouthrinse group between Group A and B (Table 3).

**Table 3.** Comparison of the $\Delta E_{00}$(Mean values±Standard deviation) $_{between}$ the groups.

| | DIS | KLO | LIS | TAN | MER |
|---|---|---|---|---|---|
| **Group A** | 0,40±0,14 | 0,55±0,23 | 1,82±2,82 | 0,50±0,24 | 1,30±0,66 |
| **Group B** | 0,34±0,15 | 0,43±0,15 | 0,96±0,66 | 0,47±0,37 | 1,15±0,27 |
| **p values** | 0,481 | 0,393 | 0,684 | 0,165 | 0,796 |

DIS, distilled water(control); KLO, Klorhex; LIS, Listerine Total Care; TAN, Tantum Verde; MER, Meridol; *p<0,05.

## DISCUSSION

According to the results of our study, MER and LIS were found to cause the most color change and thermocycling did not cause additional discoloration. The color changes caused by the mouthrinses were similar whether the thermocycling process was applied or not.

In the present study, a spectrophotometer (Spectroshade) device was used for color measurements. Spectrophotometers were proved to give reproducible, accurate, and detailed results when compared with color analyzes performed by visual methods in color measurement.[34] The SpectroShade device was also reported to have has a reliability rate of 96.9% and an accuracy rate of 80.2% within the spectrophotometers.[35]

An in vitro study revealed that Chlorhexidine®, Curacept ADS 205®, Meridol®, and Listerine Cool Citrus® did not make a statistical difference on the color stability and surface roughness of microhybrid (Point 4), bulk fill (SonicFill), and nanohybrid (Nova Compo N) resin-based composite materials, but similarly to our study, Meridol® and Listerine® caused the highest decolorization.[36] In the present study, MER was found to be different from DIS (control), KLO, TAN in group B, and DIS and TAN in group A, and it was the only mouthrinse that caused the highest color change. However, the mean value of the color change caused by the MER ($\Delta E_{00}$=1,30) was within the perceptible limits in group A and was below the perceptible limits in group B ($\Delta E_{00}$=1.15). Consistent with our results, Çelik et al. stated that Meridol® and Listerine® had the lowest pH values (Listerine pH:4,6 Meridol pH:3,88) among mouthrinses and caused more colorization than the others.[36]

In an another in vitro study that examined the effects of mouthrinses on the color stability of resin-based composite materials (nano-hybrid universal resin composite (Nova Compo N), bulk-fill resin composite (SonicFill) and polyacid modified resin composite (Dyract-XP)), Listerine Total Care Zero®, Meridol®

and Andorex® were used as mouthrinses. The results revealed perceptible color change in groups that were exposed to Meridol®, causes, but contrary with our results the least color change among the others mouthrinses was in Meridol®.[37] Composite materials were shown to have lower color stability in studies comparing the color stability of ceramic materials and composite materials.[38] The difference between our study and the other studies may be explained by the fact that all-ceramic materials have higher color stability than composite materials. Since there is no study showing the effect of Meridol® on the color change in all-ceramics, the comparisons were compared with the studies performed with composite resins. In this context, the present study offers the feature of being the first in the literature.

Several active ingredients are added to mouthrinses in order to enhance their therapeutic effects. Mouthrinses that contain fluoride for remineralization effect might effect the color change of resin composite materials, since topical fluorides were reported to cause significant changes in the color and surface roughness of different dental ceramic systems that have been glazed.[39,41] Another ingredient of alcohol reacts with cations of materials that may lead to degradation of materials, and the F anion can also be responsible for the corrosion of ceramics by interacting with Si in the structure of bioceramics.[19,20] In our study, MER with having its amine fluoride and stannous fluoride content was found to be statistically different from DIS, KLO, and TAN revealing that this condition might be due to its fluoride content.

Many studies have shown that Listerine® has a coloring effect on restorative materials.[36,42] Listerine® can cause erosion with low pH (3.5) and alcohol (21.6%) content and cause discoloration of the resin content of the material with effects such as biodegradation, dissolution of the polymer matrix.[43] In our study, LIS was found to be statistically different from DIS in both groups A and B and was the group that caused the most color change after MER.

Listerine®, Tantum Verde®, Chlorhex®, and distilled water were used as mouthrinses in the studies conducted by Soygun et al[44] where they examined the effects of mouthrinses on the color stability and surface roughness of different ceramic materials (IPS Empress CAD (Ivoclar Vivadent), IPS Emax CAD (Ivoclar Vivadent) and Lava Ultimate CAD(3M ESPE)). The samples were immersed in mouthrinses for 24 hours and 120 hours, the highest color changes were observed in the samples that were exposed to Tantum Verde® and Listerine®, respectively. The authors reported that the Lava Ultimate group, a resin nano-ceramic material, showed significant color changes in the Tantum Verde group, which may be caused by ethanol in the content.[44] Klorhex®, Tantum Verde®, Kloroben®, and Listerine® were used as mouthrinses on the discoloration of restorative materials (nano-filled composite (Filtek ultimate universal restorative resin), poly acid-modified composite resin (Dyract XP), giomer (Beautifil II) and resin-modified glass ionomer (Photac-Fil Quick Aplicap)). The highest color changes were observed in resin-modified glass ionomer samples in the Tantum Verde® group.[42] Contrarily with these studies, TAN did not cause a statistically different color change from distilled water in both of our groups. The reported coloring effect of TAN on composite materials was not observed in all-ceramic materials and did not cause color change in 1 year of use.

The coloring effect of mouthrinses that contain the active ingredient of chlorhexidine is contradictory. Highest color changes[21,22] as well as the lowest values[42,44] have been reported in various studies. Our results showed that the color change of the samples in KLO was not statistically different from DIS and the mean color change values were below the perceptible limits.

The effect of three existing aging protocols on the color change of all- ceramic systems were investigated and the color changes of samples prepared in two different thicknesses (0,5 and 1 mm) were compared with accelerated artificial aging, autoclave aging and thermocycling methods. Color changes were reported to be different according to the aging method and the 0.5 mm thick samples showed more color change than the 1 mm thick samples.[45] In the present study, the samples aged with 10,000 cycles by thermocycling did not show a statistically significant color change and the samples prepared with a thickness of 1,5 mm did not show any color. The results confirmed the main hypothesis of the current study that mouthwashes cause color changes in all ceramic materials, but showed that thermal cycling did not make any significant changes.

The limitations of our study are that the in vitro conditions in our study cannot fully mimic the oral environment and the aging method used in the study has the possibility of revealing different results in terms of color change values. It is also known that color changes can be affected by surface finishes, material type and thickness. Other factors that could influence the color changes like, diet or abrasion were not included. The results should be interpretted by additional in vivo and in vitro studies.

## CONCLUSION

Within the limitations of the study, MER and LIS were shown to cause color change in all-ceramic materials. Clinicians should consider the possibility of staining when recommending MER and LIS for routine use to their patients, particularly when planning all-ceramic restorations thinner than 1.5 mm.

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**Conflict of Interest**

The authors declare no conflict of interest.

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