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In-vivo evaluation of the effects of short-term dehydration and rehydration on the color differences of immature permanent incisor teeth

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Abstract

Objective: It is aimed to evaluate the color changes of upper central incisor teeth with different root maturation levels after 15 min of dehydration and rehydration.

Materials and Methods: Using the Cvek classification, 60 participants were divided into five groups. A spectrophotometer, equipped with a specially designed jig, was utilized for color measurements. Following isolation, color measurements were conducted at the dehydration process intervals of baseline, 1st, 2nd, 3rd, 5th, 7th, 10th, and 15th minutes. After mouth rinsing, measurements were repeated at the same time intervals during the rehydration process. CIEDE2000 (ΔE_{00}) values and the Whiteness Index (WI_D) were obtained to compare the color changes between the initial readings and each interval. Friedman and Repeated Measures ANOVA were applied to compare classes by time during the dehydration and rehydration periods, as well as ANOVA and Kruskal-Wallis tests to compare by classes for each time ($p < 0.05$).

Results: Significant differences emerged in the mean ΔE_{00} values during dehydration and rehydration periods across all Cvek Classes ($p < 0.05$). While no statistically significant differences were observed among Classes at different minutes during the rehydration period ($p > 0.05$), it was particularly noted that the mean ΔE_{00} values of Classes 2, 3, and 5 at the 2nd minute of dehydration were statistically significantly higher than the mean of Class 1 ($p < 0.05$). However, when evaluating the starting and ending measurements of both dehydration and rehydration processes, and the overall change in ΔE_{00} and ΔWI_D , no significant variations were detected across the Classes ($p > 0.05$).

Conclusions: In teeth that have just erupted and are still undergoing root development, dehydration in the initial minutes results in noticeable color changes. However, the change in ΔE_{00} and ΔWI_D of the teeth was found to be similar after the dehydration and rehydration processes.

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Clinical Significance: According to the Cvek classification, the lower color change in Class 1 teeth within the first 2 min of dehydration and the need for more than 15 min of rehydration for immature teeth with an open apex to return to their initial state after a 15-min dehydration are crucial for clinicians in terms of color assessment timing.

KEYWORDS

color change, immature teeth, open apex, operative dentistry, teeth dehydration, teeth rehydration

1 | INTRODUCTION

When teeth begin to be consistently exposed to air, such as during dental treatment procedures, the moisture on their surface is lost, leading to dehydration. This increases the opacity of the enamel, giving teeth a whiter appearance.¹ As the spaces between the prisms are water-filled, the enamel layer appears semi-transparent, allowing light to disperse between the prisms. In contrast, dentin is richer in color and chroma.¹ Dehydration causes these spaces to fill with air rather than water. This change in transmitting medium results in different light refraction patterns, making the enamel less transparent and brighter.^{2,3} Consequently, the dentin's color is masked, making teeth appear lighter or more opaque. The organic components in dentin predominantly absorb light in the ultraviolet and visible spectral range.⁴ Furthermore, factors like the orientation, number, and size of dentin tubules can influence light scattering.⁵ The light's transmission can be impacted by the thickness of both the dentin and enamel, and by the enamel's transparency.⁶

The interaction of color perception and matching skills hinges on ideal circumstances, where light reflecting off an object from a specific source is observed. This interplay involves three key elements: the characteristics of the light source, the optical properties of the observed object, and the observer's aptitude for interpreting colors.⁷ The operator's proficiency in color matching is impacted by factors such as viewing conditions, eye fatigue, age, experience, and the presence or absence of color vision deficiencies.⁸

There are two primary methods to assess tooth color clinically: subjective visual determination using tooth shade guides and employing spectrometers. Spectrophotometers, spectroradiometers, and colorimeters are instruments used in such measurements.^{9–17} Visual color selection using a guide is a quick and cost-effective method, but it is considered inconsistent and subjective. Many factors, such as proper lighting, color acuity, and eye fatigue, can affect this color selection method. Using advanced tools can help overcome these undesirable conditions.¹⁸ However, even if the latest technology is used, if sufficient attention is not paid to the basic principles of color selection, undesired color mismatches can emerge in the final restoration.

Root development initiates after enamel and dentin formation is complete in the tooth crown. The inner and outer enamel epithelia proliferate, merging to form Hertwig's epithelial root sheath, a bilayered structure resembling a tube, responsible for creating the

tooth's root or roots. Subsequently, it undergoes fragmentation and resorption, shaping the root.¹⁹ As the sheath nears the root's apical region, it narrows to create the apical foramen. Root development persists even after tooth eruption, typically concluding within an average of 3 years post-eruption. An additional concern arises from the ongoing development of dental enamel post-eruption.²⁰ Variances established during the initial stages may account for the observed outcomes in teeth where external mineralization has not promptly concluded. Complicated and uncomplicated crown fractures constitute the most common form of dental trauma in permanent teeth, especially in the early mixed dentition period when the apical opening has not yet fully closed.²¹ The teeth most commonly affected by this condition are the permanent maxillary central incisors.²² In this context, determining tooth color during esthetic rehabilitation requires being fast and practical, including dehydration and rehydration processes.

During this period, it has been reported that the blood circulation in the tooth pulp changes over time and that teeth with closed apices have lower pulp oxygen saturations.²³ In light of this information, it has been anticipated that enamel/dentin dehydration might differ based on the changes in blood circulation within the tooth (pulp). In the literature, color changes in teeth with completed root development after dehydration, and their rehydration processes have been evaluated based on different parameters.^{9–16} As far as we know, there is no research on how the dehydration-rehydration process progresses at different root development stages of teeth with open apices, how the optical properties of the tooth crown change, and how they compare with teeth with closed apices.

This study's objective is to determine how root apex opening levels impact color parameters during dehydration and rehydration processes in immature permanent upper incisor teeth with different level unclosed apices. This is done by considering the relationship between pulp blood flow and the pulp-dentin-enamel interaction. The null hypothesis of the study is formulated on the premise that the level of root apex openness will not have an effect on the color parameters during the dehydration and rehydration periods.

2 | MATERIALS AND METHODS

Clinical Research Ethics Board of the Çanakkale Onsekiz Mart University School of Medicine granted ethical approval for this research

(Decision no = 2023/04-05). The study adhered to all principles outlined in the Declaration of Helsinki.

2.1 | Sample size calculation

G*Power 3.0.10 statistical software (v.3.1.9.7, Universitat Kiel, Kiel, Germany) was used to calculate sample size. Based on the data from one study¹⁴ which showed that the Delta E (ΔE_{00}) values of the 10 min in dehydration process 2.2 ± 1.6 compared to the 2 min in same process which was 1.1 ± 1.0 in the central incisors, a total sample size of 52 was calculated to be enough to achieve power of 90% with a significance level (α) set at 0.05. Considering the 10% dropout risk, the minimum sample size of the study was determined as 57. Since there were 5 groups in total in the study and there should be an equal number of teeth in each group, the number of samples was determined as 12 in each group, a total of 60.

2.2 | Participant selection

Participants were selected based on specific criteria from among patients who sought treatment at the Pediatric Dentistry department of Çanakkale Onsekiz Mart University Faculty of Dentistry between March and August 2023. Radiological evaluations and the selection of appropriate patients were conducted by Berkant Sezer, a specialist in Pediatric Dentistry with expertise in these areas and publications on these topics. The permanent central incisor teeth of the patients who applied to the clinic for routine treatment and required panoramic radiographs for diagnosis/treatment were evaluated and classified radiographically. Using a Promax 2D (Planmeca Oy, Helsinki, Finland) with settings of 60–64 kV, 5 mA, and an exposure time of 14 s, radiographs were captured. The quality of the radiograph is graded into three categories based on its clinical significance, as outlined by the UK National Radiological Protection Board (NRPB) criteria²⁴: excellent, diagnostically acceptable, and unacceptable. Grade 1 radiographs are categorized as excellent and are defined by the absence of errors in patient preparation, positioning, exposure, rendering, or film processing. To ensure the highest level of evaluation accuracy, our study exclusively assessed these grade 1 radiographs.

2.2.1 | Inclusion criteria

- The participants do not have any adverse health conditions and are not taking any medications. Unstimulated saliva flow has been measured in terms of volume to ensure it is within normal limits (0.3–0.4 mL/min).
- The tooth to be evaluated (central incisor) should be a natural tooth with healthy dental tissues and periodontal status, free from restorations, wear, severe color changes, and hypoplasia or hypomineralization.

2.2.2 | Exclusion criteria

- Participants with adverse health conditions and those taking medications. Presence of restorations, caries (including white spot lesions), and hypoplasia or hypomineralization;
- Currently undergoing orthodontic treatment or have received it in the past.
- Presence of fixed orthodontic appliances, including central incisors.
- Dry mouth or low levels of unstimulated saliva flow.
- Conditions where tooth integrity is compromised such as fractures, cavities, restorations, and histopathological conditions in enamel-dentin tissues.
- Cases where the tooth crown has not fully erupted.
- Having previously undergone whitening or resin infiltration on the anterior teeth.
- Patients who cannot breathe through their nose.

After the initial radiological assessment, 80 patients were identified, and 20 patients were excluded for various reasons. The remaining 60 patients were divided into five classes, forming subgroups consisting of 12 patients each ($n = 12$). Following a radiological evaluation of the 60 participants who met the inclusion criteria was carried out by Pediatric Dentistry specialist Berkant Sezer, they were divided into five subgroups using Cvek's root apex maturation classification²⁵:

- Class I = Less than half the root length.
- Class II = Half of the root length.
- Class III = Between half and two-thirds of the root length.
- Class IV = Almost full root length with a wide-open apical foramen.
- Class V = Closed apical foramen with completed root development.

Radiographic examples of the permanent incisors of the patients included in the study according to the relevant classification are given in Figure 1.

2.3 | Color measurements

Before the measurements, all participants were requested to brush their teeth without using toothpaste for a duration of 2 min. All participants were seated and reclined in the dental unit positioned at a 45° angle relative to the ground to eliminate shading effects from the nose or lips. Color measurements were conducted under lighting conditions where ambient light sources remained constant (with a color temperature of 5500 K and a Color Rendering Index [CRI] of 95). Lips were retracted using a retractor (Ocean Aquarius Mouth Opener for Speak Out Game C-Shape Lip Retractor) and cotton rolls. The placement of the retractor and cotton was completed within 15 s; otherwise, the patient was allowed to close their mouth, waited for a few minutes, and the process was repeated to ensure there was no pre-dehydration. Separate clear silicone guides were made for each patient to help with measurements in the central third of the tooth

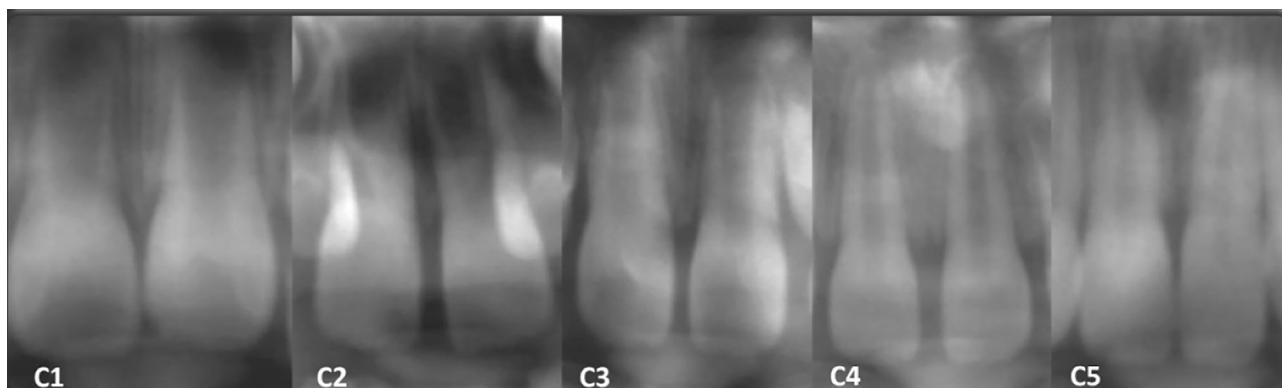


FIGURE 1 Radiographic classification of the upper permanent incisors of the patients. (C: Class).

where measurements will be taken. A standardization of assessments suitable for both the patient and the dentist was planned for ergonomic measurements, with a preference given to prioritizing tooth number 11 in the initial evaluation. A silicone guide was placed, and before each measurement, three readings were taken from the designated upper central incisor tooth's middle of facial surface at each time interval using the calibrated VITA Easyshade 5 Spectrophotometer (VITA Zahnfabrik, Bad Säckingen, Germany).^{26,27} The measurements were conducted in a consistently standardized clinical environment with controlled temperature and humidity, at the same location and time of day.

In the CIEDE2000 system, Delta E (ΔE_{00}) gauges the overall color difference between two color samples, taking into account differences in Lightness (L), Chroma (C), and Hue (H).^{28,29} The CIEDE2000 color-difference formula includes specific corrections for CIELAB's non-uniformity (the so-called weighting functions: S_L , S_C , and S_H), a rotation term (R_T) that accounts for the interaction between chroma and hue differences in the blue region, a modification of CIELAB's a^* coordinate, which primarily affects colors with low chroma, as well as parameters accounting for the impact of lighting and vision conditions (the so-called parametric factors: K_L , K_C , K_H).²⁸ K_L , K_C , and K_H are recommended to be set as 2:1:1 in dental research and instrumental color analysis.³⁰ After three-stage calculations using the L^* , a^* , b^* values obtained with the Vita EasyShade device, the data symbolized in the equation below were determined, and calculations were carried out to calculate ΔE_{00} values:

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

Measurements were taken at time intervals of 0 (initial), 1st, 2nd, 3rd, 5th, 7th, 10th, and 15th minutes. All participants were asked to use the same breathing method to ensure similar air flow affecting the tooth, specifically to breathe through their nose during the measurements. Following the dehydration procedure, the lip retractor and cotton rolls were taken out, and a comprehensive mouth rinse with water was performed for a duration of 30 s. The rehydration of the

teeth commenced upon contact with saliva. Participants were instructed to consume a glass of water (200 mL) and keep their mouths closed for a period of 5 min. After the 5-min interval, measurements were taken again at the baseline, 1st, 2nd, 3rd, 5th, 7th, 10th, and 15th minutes using the same silicone guide. Patients only opened their mouths during the measurements and kept them closed between measurements. ΔE_{00} values were compared with acceptability threshold values ($\Delta E_{00} = 1.8$) and perceptibility threshold values ($\Delta E_{00} = 0.8$).³¹ Additionally, CIE Lab* (CIELAB) color parameters (L^* , a^* , b^*) results were recorded for comparison with previous studies.

Recently, a newly developed approach has introduced a specialized dentistry Whiteness Index based on the CIELAB color space, referred to as WI_D .³² Participants' WI_D data at each measurement during dehydration and rehydration stages were calculated using the formula ($WI_D = 0.511 L^* - 2.324a^* - 1.100b^*$).³³ The differences in WI_D between the initial measurement of dehydration and the ongoing measurement minutes during dehydration and rehydration were measured using the equation $\Delta WI_D = |WI_{D2} - WI_{D1}|$, and the changes in WI_D values over the course of the experiment relative to the initial measurement were assessed. In the assessment of ΔWI_D , the perceptibility threshold (PT) has been set at 0.72, and the acceptability threshold (AT) at 2.62 units.³³

2.4 | Statistical analysis

In the study, descriptive statistics for the data (mean, standard deviation, minimum, maximum, median, and confidence interval) were provided. Normality assumption was checked with the Shapiro Wilk test and variance homogeneity with the Levene test. Friedman and Repeated Measures ANOVA (Greenhouse–Geisser Statistics) were applied to compare each class by time during the dehydration and rehydration periods, as well as ANOVA and Kruskal–Wallis tests to compare classes within each time interval. For multiple comparisons, Bonferroni Post Hoc test was performed. Analyses were conducted in the IBM SPSS 25 program and p -values of less than 0.05 were considered to indicate statistical significance.

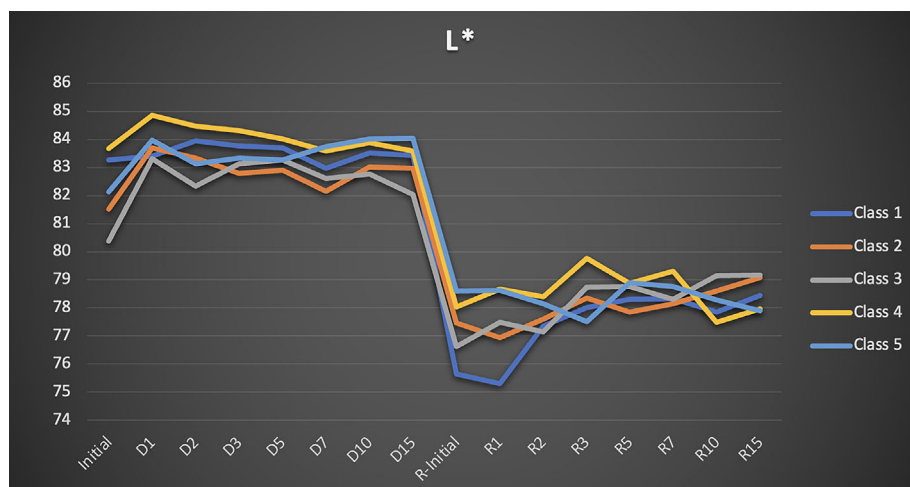


FIGURE 2 Line graph of L^* measurements between Classes over time during the dehydration and rehydration periods.

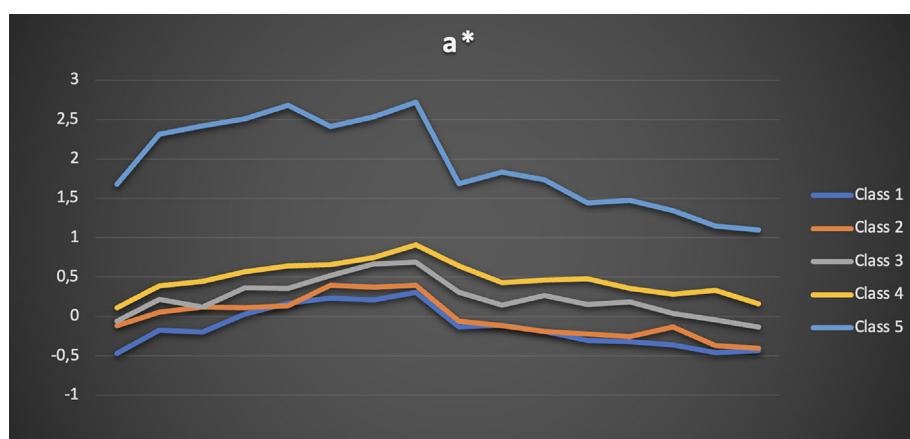


FIGURE 3 Line graph of a^* measurements between Classes over time during the dehydration and rehydration periods.

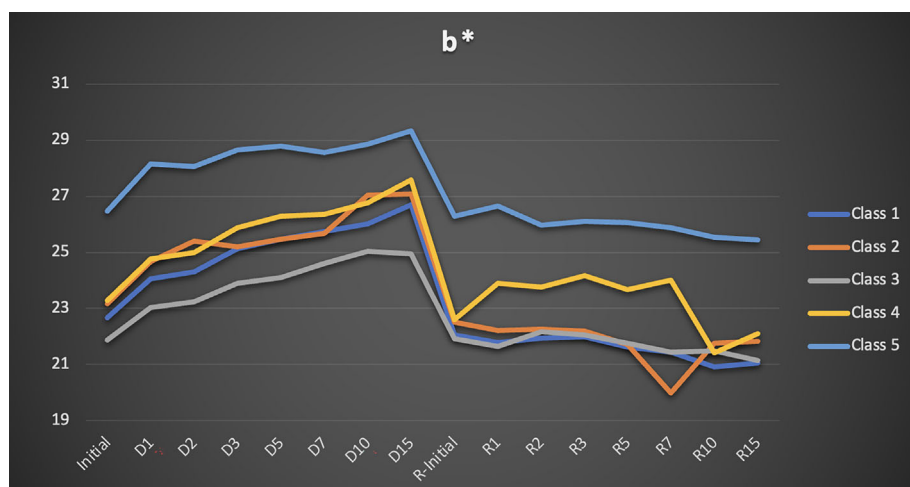


FIGURE 4 Line graph of b^* measurements between Classes over time during the dehydration and rehydration periods.

3 | RESULTS

Among the participants included in the study, 35 were males while 25 were females. The mean age ($\pm$ standard deviation) of the evaluated patients in the study is 8.6 ± 0.8 has been found to assess the homogeneity of the age distribution.

The mean values of the L^* , a^* , b^* parameters of teeth were also determined according to Classes (Figures 2–4). In numerical terms, the mean values of the a^* and b^* parameters in Class 5 patients were higher at all measurement times compared to the teeth of other Classes (Figures 3 and 4). The ΔWID results are as shown in the Figure 5. In all Classes, statistically significant differences were found between

FIGURE 5 Line graph of ΔWI_D measurements between Classes over time during the dehydration and rehydration periods.

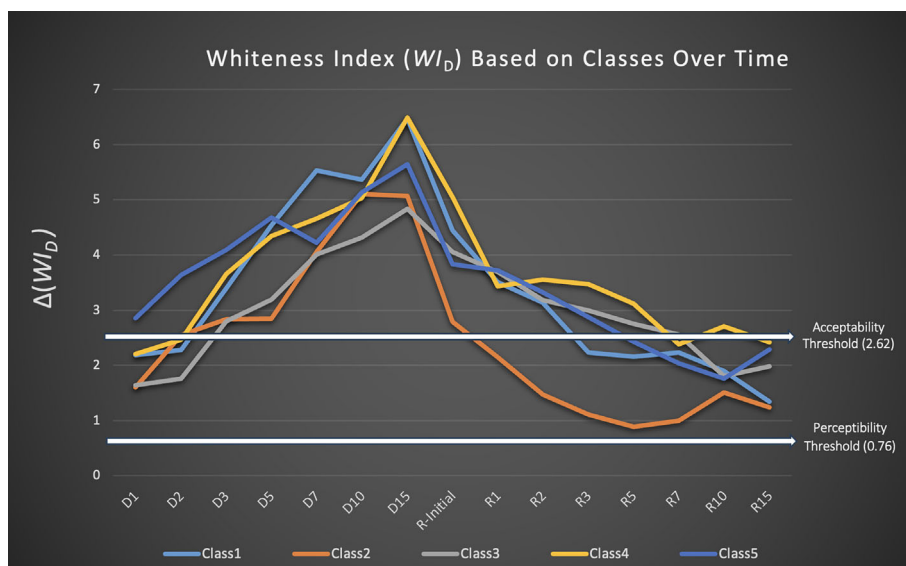
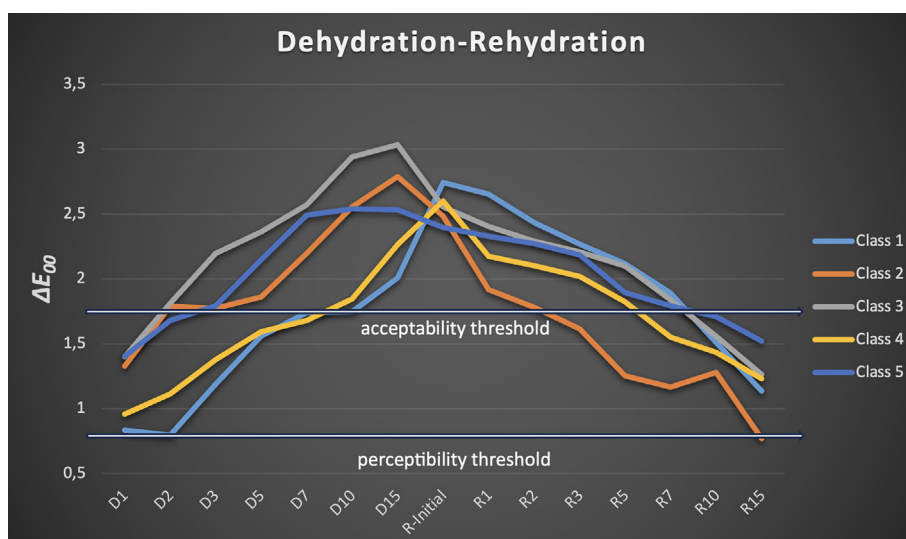


FIGURE 6 Line graph of ΔE_{00} measurements between Classes over time during the dehydration and rehydration periods.



the within-group ΔWI_D measurement values calculated during dehydration and rehydration periods ($p < 0.05$). In the analysis comparing ΔWI_D values among Classes at different measurement times, only the inter-class difference at the 5th minute of the rehydration period was statistically significant ($p = 0.027$).

Based on the ΔE_{00} values taken at the 1st minute, we observed that the percentages of results exceeding the unacceptable limit ($\Delta E_{00} = 1.8$), when ranked from Class 1 to 5, were 16%, 22%, 22%, 10%, and 22%, respectively. At the 2nd minute, the percentages were again measured as 16%, 22%, 44%, 10%, and 33% in order of classification. Considering the study as a whole, only 23% of the teeth exceeded the acceptable limit at the 2nd minute based on measurements from all classes. Additionally, within each group, the minutes at which the number of teeth surpassing the acceptable threshold ($\Delta E_{00} = 1.8$) began to represent more than half of the group are reported as follows, in the same sequence: 15th, 10th, 5th, 7th, and 5th minute (Figure 6).

Statistically significant differences were found between the mean ΔE_{00} values over time during the dehydration and rehydration periods in all Classes ($p < 0.05$). For each time parameter based on Classes, a statistically significant difference was found between the mean measurements of the 2nd minute during the dehydration period ($p < 0.05$). According to the Bonferroni tests, statistically significant differences were determined between Class 1 and Class 2, 3, and 5 groups ($p = 0.023$, $p = 0.008$, and $p = 0.007$). The mean ΔE_{00} values of Class 2, 3, and 5 groups are higher than the mean of the Class 1 group. Moreover, no statistically significant differences were found between the mean ΔE_{00} measurements of the 1st, 3rd, 5th, 7th, 10th, and 15th minutes of the dehydration period and all rehydration periods across Classes ($p > 0.05$).

For the Classes, the beginning and end measurements of the dehydration and rehydration periods, as well as the change in ΔE_{00} from the beginning of dehydration to the end of rehydration, were analyzed. No statistically significant differences were found in the

TABLE 1 Comparison of the coloration (ΔE_{00}) measurement changes for groups based on classes.

Group	Time (min)	Class	Mean $\pm$ SD	Median	Min-Max	95% C.I. (Upp-Low)	Test statistics	<i>p</i>
Deh.	15th-1st	Class 1	1.2 $\pm$ 0.5	1.2	0.4-2.4	0.9-1.6	4.704***	0.319
		Class 2	1.4 $\pm$ 0.6	1.3	0.6-2.8	0.9-1.9		
		Class 3	1.6 $\pm$ 0.9	1.5	0.1-3.0	0.9-2.3		
		Class 4	1.3 $\pm$ 0.5	1.2	0.6-2.2	0.9-1.6		
		Class 5	1.1 $\pm$ 0.9	1.0	0.4-3.4	0.4-1.8		
Reh.	15th-Int.	Class 1	1.8 $\pm$ 0.8	1.6	0.6-3.4	1.2-2.3	3.267***	0.514
		Class 2	1.7 $\pm$ 1.3	1.7	0.0-4.6	0.6-2.7		
		Class 3	1.4 $\pm$ 1.1	0.9	0.3-2.9	0.5-2.2		
		Class 4	1.3 $\pm$ 0.8	1.3	0.2-2.7	0.7-1.9		
		Class 5	1.1 $\pm$ 0.5	5	0.4-2.0	0.7-1.5		
Reh-Deh	Rh15-Dh.1	Class 1	0.9 $\pm$ 0.8	1.0	0.2-2.1	0.4-1.4	3.890	0.421
		Class 2	1 $\pm$ 1.1	0.6	0.1-3.5	0.1-1.8		
		Class 3	0.7 $\pm$ 0.7	0.4	0.0-2.1	0.2-1.2		
		Class 4	1.0 $\pm$ 0.7	0.4	0.0-2.0	0.5-1.5		
		Class 5	1.4 $\pm$ 0.9	0.9	0.2-2.7	0.7-2.1		

***Kruskal-Wallis test.

mean changes of ΔE_{00} measurements across the classes ($p > 0.05$), as indicated in Table 1. In all Classes in the study, the mean ΔE_{00} data between the final measurement after 15 min of rehydration and the initial measurements of dehydration were found to be less than 1.8 (Table 1). It is observed that the teeth in Class 1 exceed the unacceptable ΔE_{00} limit later compared to the closed apex group (Class 5) and remain at this level for a shorter period of time.

4 | DISCUSSION

Based on the current data, our null hypothesis suggesting that the classifications made according to dental root development and apex opening would not have an effect on color changes during dehydration and rehydration processes has been rejected. Furthermore, in the 2nd minute of the dehydration period, the ΔE_{00} values of the teeth in the Class 1 group being lower than those of the other groups indicate that teeth with open apices are exposed to color changes more slowly compared to teeth with closed or semi-closed apices in the earliest stage. However, as the minutes progress, this situation becomes equivalent with the other groups, and the color change graph follows the same trend as the other groups.

L^* - Lightness coordinate ($L^* = 0$ represents black, and $L^* = 100$ represents white), a^* - Red/Green coordinate, $+a^*$ indicates red, and $-a^*$ indicates green, b^* - Yellow/Blue coordinate, $+b^*$ indicates yellow, and $-b^*$ indicates blue.³ When compared to other similar studies, lower a^* values and higher b^* and L^* values were reported in the measurements of this study.^{13,14} In both compared studies, central teeth were examined. However, the mean age of the study group is higher than the age group in this study. In another recent study, it has been

shown that the a^* and b^* values increase with age in older patients compared to younger ones.¹⁶ This result is consistent with our study. Participants in Class 5 have higher a^* and b^* values in all measurements compared to others. Furthermore, when comparing the L^* values of participants, it is reported once again that the L^* value decreases with age, yielding results similar to the literature and recent studies.^{12,13,16}

The differences in L^* , a^* , b^* values are also reflected in the results of ΔWI_D . In dehydration studies where participants with a higher mean age than our study are evaluated, the ΔWI_D in maxillary central incisors teeth increases with the duration of dehydration.¹⁴⁻¹⁶ This study also has yielded results parallel to similar studies in the literature.^{14,16} As the dehydration period progresses from the beginning to the end, an increase in this value is observed, while a decrease in ΔWI_D values is noted during the rehydration process. All evaluated teeth had reached acceptable ΔWI_D values at the end of the rehydration process, but none of them had fallen below the perceptibility threshold. Similarly, in the study evaluating ΔWI_D values depending on age changes, we can observe differences in the ΔWI_D profile as age progresses.¹⁶ The ΔWI_D profile of patients aged between 20 and 30 and those aged between 65 and 80 exhibits variations. It is presumed that in our study, with a relatively narrow age range, the ΔWI_D graph resulted in similar outcomes. It is anticipated that these differences may arise with age, in conjunction with tooth maturation (continuity of dentin formation, ongoing enamel mineralization, external factors such as staining agents, etc.).¹⁷

For patients in the Class 1 group, the mean time at which their ΔE_{00} measurements exceeded the acceptability threshold limit was 7.7 min from the start, and the mean duration they remained above this value was 13.4 min. The same situation was observed in the closed apex Class 5 group patients, where the mean time for the ΔE_{00}

values to exceed acceptability threshold limit was 4.6 min, and the duration they remained above this limit was found to be 18.8 min. While the rejection of the hypothesis is based on statistical results, it is noteworthy in the mean outcomes that the Class 1 teeth reached the unacceptable ΔE_{00} limit later compared to Class 5 teeth and remained in this state for a shorter duration.

In the literature, while studies on similar topics are limited, no study has been encountered yet that uses a similar experimental method based on the degree of root development. One study reported that teeth returned to their initial values 30 min after being isolated with a rubber dam for 15 min.¹¹ On the other hand, after 15 and 30 min of rehydration, the color differences were notable in teeth that underwent dehydration for both 15 and 30 min.^{10,12} In another study, the mean color difference after 24 h of dehydration was beneath the perceptibility threshold compared to the baseline.¹³ In the context of our research, subsequent to a 15-min rehydration process, none of the teeth reverted to their original color values. In this context, for this study within the age group (7–12), the measurement time needs to be extended in future studies to determine when the tooth returns to its own color parameters after 15 min of dehydration.

In terms of acceptability from another study, over 95% of the teeth examined exhibited color variations beyond the acceptability threshold after 10 min of dehydration, while nearly 70% displayed unacceptable color differences just after 2 min of dehydration.¹⁴ In this study, this situation varies by groups and is quite different. Based on the data from our study, it can be stated that newly erupted teeth surpass the acceptable ΔE_{00} limits after longer durations of dehydration onset compared to the data obtained from older teeth reported in the literature.

In a recent study involving a sample of 30 participants, a color difference of $\Delta E_{00} = 2.02$ units was observed between the baseline and 10 min of dehydration for both central and lateral incisors.¹³ Additionally, after 10 min of dehydration, 95.8% of the teeth examined exhibited ΔE_{00} values above the perceptibility threshold, and 54.1% exceeded the acceptability threshold, consistent with the reported percentages.¹⁴ In this study after 15 min of dehydration, the distribution percentages of teeth with a ΔE_{00} value over acceptability threshold ($\Delta E_{00} = 1.8$) compared to the patients within the group, when listed from Class 1 to Class 5, are reported as 50%, 88%, 88%, 80%, and 88%. After 15 min of rehydration, 19.2% of all the patients evaluated in the study had ΔE_{00} values above the acceptability threshold limit, while almost half of the participants (40.3%) were observed to have values below perceptibility threshold ($\Delta E_{00} = 0.8$).

Methodological differences create challenges in directly comparing the results of the study with other research. Participant numbers in the studies, their presence in different age groups, color measurements from different teeth, the use of different color measurement devices, and the limited number of in-vivo studies are among the other variations in this study.^{9–17}

Another issue is the continued maturation of dental enamel after its eruption. Differences formed in the early phase can be anticipated

as a reason for the present results for teeth whose external mineralization has not been completed immediately.²⁰ The increased hydratability of enamel prisms that have not yet completed mineralization compared to mature enamel, as well as the impact of this situation on the speed of dehydration and rehydration, is a subject that needs to be examined in further research. It has been reported that the blood circulation in the dental pulp is lower in teeth with closed apices, leading to lower pulp oxygen saturations.²³ It is thought that dehydration of enamel/dentin may vary based on changes in pulp blood circulation.

Color measurements were conducted during the dehydration and rehydration stages in this study. Consequently, utilizing a rubber dam wasn't feasible. Thus, patients were instructed to keep their mouths open while the color measurements were taken. In prior studies, color measurements were carried out using a VITA Easyshade 5 (VITA Zahnfabrik, Bad Sackingen, Germany).^{11–13} An in vitro study reported that the VITA Easyshade had a repeatability of 96.4%, in comparison to the 96.9% of the SpectroShade; their accuracy rates were documented at 92.6% and 80.2% respectively.²⁶ In a clinical study, the SpectroShade demonstrated superior repeatability in comparison to the VITA Easyshade.²⁷ This aspect can be viewed as one of the study's limitations.

The limited number of current patients, measurements taken from only one tooth, and the type of device used for color measurements are the prominent limitations of the study. A limitation related to the method is that even though the oral cavity is considered black, small differences in the background color can affect color readings, hence a standardized background could not be used. Our imaging-related limitation is that we used the panoramic radiography method instead of the periapical radiography technique to classify the root development stage and apical opening. In line with ethical principles and obligations, we did not perform additional imaging. This study, as far as we know, will be effective in that it is the study that evaluates and reports color change parameters due to dehydration on immature teeth, and will also be a pioneer in further research on the same subject in this age group. Available data will contribute to clinicians. For future research, for studies in this age group, it would be beneficial to; conduct the study on a larger number of patients and teeth, evaluate the other teeth in addition to the central incisor teeth, examine the changes in dehydration at shorter time intervals, determine with more precision the moment when the perceptibility and acceptability thresholds are exceeded using spectroradiometers, which are considered the gold standard.

5 | CONCLUSION

Within the limitations of this current study, it was concluded that:

1. According to the Cvek classification, the color change in ΔE_{00} value due to dehydration for the teeth in Class 1 is lower in the first 2 min compared to other groups.

2. The changes in color (ΔE_{00}) values and the Whiteness Index ($\Delta W I_D$) values of the teeth was found to be similar during the dehydration and rehydration processes; moreover, the values between the initial measurements of dehydration and the final measurements of rehydration are also similar.
3. After 15 min of dehydration, the distribution percentages of teeth exceeding the acceptability threshold ($\Delta E_{00} = 1.8$) compared to the patients within the group, when listed from Class 1 to Class 5, are reported as 50%, 88%, 88%, 80%, and 88%.
4. In all groups, teeth required more than 15 min of rehydration to regain their original shade after 15 min of dehydration.

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CONFLICT OF INTEREST STATEMENT

The authors have no financial interest in any of the companies whose products were used in this study.

DATA AVAILABILITY STATEMENT

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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